

# Unify Toxicity Module

## ECOTOXicology Database System

### ECOTOX Coding Guidelines

Prepared for:

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# REVIEW OF LITERATURE

## A. OVERVIEW OF ECOTOX

### 1. Introduction to Review of Literature

The data elements included in ECOTOX encompass standard test parameters typically reported within a publication. Each database record contains information about the exposure and test conditions. Specific parameters include the test chemical, species, and endpoint or effect concentration.

The included literature is identified through standardized bibliographic retrievals. Each publication is evaluated and the applicable data is encoded by trained literature reviewers. The data encoded are evaluated according to existing standard test methods such as those from the American Society for Testing and Materials (1996), Code of Federal Regulations (1992), and the American Public Health Association (1992). Each test included in ECOTOX is assigned a documentation code that indicates the amount of supporting methods and results documentation available in the original scientific publication.

**Note:** Each publication included into the ECOTOX database must meet the five minimal criteria for acceptance (i.e. chemical, species, concentration, duration and effect) . If the paper is missing one or more of these criteria ECOTOX does not search other sources to obtain the missing data piece(s). Sources such as author communications and referencing another work to obtain one of the five criteria is allowed for specific risk assessment/criteria projects (e.g. EcoSSL or CAD) and must be specified by the EPA Database Coordinator.

### 2. Literature Reviewer Training

#### Training Sequence

The training sequence is designed to develop consistent, accurate, and versatile literature reviewers. This is accomplished through an intensive period of literature review, interactive quality assurance procedures, and consultation with other ECOTOX database personnel.

The scope of the six month intensive training period encompasses the following areas:

- endpoint toxicity test review (one month);
- effect only toxicity test review (two months);
- bioconcentration study review (one month);
- field study review (one month); and in-depth training within the areas listed above (one month).

The personnel available to support the reviewer include the data coordinator and trained ECOTOX reviewers. The following documentation and materials are used for training:

- ECOTOX Standard Operating Procedures (<http://www.epa.gov/ecotox> under "download files");

- Fundamentals of Aquatic Toxicology (Rand (Ed), 1995);
- American Society for Testing and Methods; (ASTM, 1996);
- Selected toxicity literature publications

The reviewer initially reviews the ECOTOX Standard Operating Procedures: Coding Guidelines, applicable publications listed in the reference section for each of the databases, applicable US EPA Standard Evaluation Procedures and ASTM guidelines. The primary emphasis is to understand the minimum criteria that characterize acceptable toxicity tests. These criteria must be reported in the toxicity publications selected for review in order to qualify for inclusion in the ECOTOX database.

The acceptance criteria are:

- Name of the test **chemical**;
- Name of the **test organism**;
- **Effect** of the test chemical on the organism;
- Test chemical **concentration** or application rate;
- Test **duration** (except for abstracts and non-English publications).

The secondary emphasis is to develop the ability to distinguish between exposure types (lethal, sublethal, bioconcentration). The reviewer is trained to recognize whether standard methods are reported for test methodologies and for the test endpoint. The reviewer is also trained to identify tests which are not applicable to ECOTOX.

Once the general introductory materials are read, the standard training guidelines introduce the reviewer to each category of toxicity literature. Information specific to areas of acute, chronic and bioconcentration literature is discussed in subsequent sections of this chapter. The guidelines can be tailored to the specific areas of expertise and strengths that each person brings to the project. Three primary elements are emphasized in each component of the training sequence. The standard training sequence is:

1. Example review: Examination of previously encoded toxicity literature. The trainee reviews between 5 to 10 toxicity publications and compares each with its associated pre-completed coding printout.
2. Independent review: The trainee independently reviews a minimum of 10 to 20 toxicity publications. All 10 - 20 reviews are quality assured via a review of the publication and coding by the data coordinator. Inconsistent coding practices are resolved with the trainee. The trainee continues to review additional toxicity publications and the level of QA decreases from 100 percent to 10 percent as the reviewer's consistency and proficiency increase.
3. Measure of proficiency: Established ECOTOX quality assurance procedures require a close review of all reviewed publications by the data coordinator to ensure accurate reviewing is consistent with current test methodologies and SOPs. All discrepancies identified are noted by the data coordinator and discussed with the trainee.

4. A full time reviewer begins the training sequence reviewing 20 publications per month. This amount increases until a level of 35 publications per month is attained. The average time estimated per review at the beginning of the training sequence is 1.5 hours per publication. The time should decrease to one hour per publication. A part time reviewer's training expectations will be decreased accordingly.

#### Measures of Competency for Trained Reviewers

The quality assurance process is an ongoing component of literature reviewing. Emphasis is placed on quality assurance during the initial collaborative training period, during the 10 percent replicate review process, and through consultation with publications in the field of aquatic toxicology. As part of this process, consistency and concurrence between the document abstractors is attained.

The ten percent replicate review process assures data integrity and promotes routine evaluation of coding practices. Through this training process, strengths and weaknesses in the data abstractor's expertise are identified and specific programs are established to enhance expertise where needed. Such programs include consultation with ECOTOX staff, toxicology publications and the EPA Database Manager, as needed. Evaluation of replicate reviews, which is performed on 10% of all coded references, is used to flag and correct any major discrepancies between replicates. In addition a screening of all completed entries to ensure consistency and completeness prior to data entry is required. Parameters routinely screened include water chemistry, test organism descriptors, calculated endpoints and total test numbers.

#### Steps in the Quality Assurance Process

1. Ten percent of the reviewed articles from each abstractor are randomly identified by the data coordinator. Information concerning the number of publications is entered into a file, maintained on the data coordinator's computer. The spreadsheet tracks the QA process and calculates the percent of the publications subjected to quality assurance for each reviewer (Table 1). The original reviewer's entry printout for the chosen publication is placed in the "Double Review and QA" file folder maintained by the data coordinator. An "ECOTOX 10% Tracking Form" sheet is maintained in the folder and filled out as articles are received (Attachment 1.). The spreadsheet file is also updated.



**Table 1. ECOTOX 10% TRACKING SHEET EXAMPLE**

Date Rec	Doc #	Tot Rec'd	# QAed	2nd Rev	2nd Comp	Coord	Completed
11/30/93	5342	8	1	JACKY	11/30/93	ANNE	01/15/94
12/14/93	6808	10	1	AMY			

2. The publication is given to a second reviewer for independent review. After completion of the second review, the data coordinator gives the copies of entry sheets and paper to the EPA Database Manager who compares both reviewer's entry printouts, documents the differences (if any) between reviewers, archives the information on the "ECOTOX 10% Replicate Review" form (Attachment 1.), then returns the form to the reviewers for comment. The reviewers note discrepancies by either agreeing with the EPA Database Manager's comments or expressing their differing opinions on the form. After the replicate review form is returned to the EPA Database Manager, discussions are held with both reviewers to resolve any remaining differences. Discrepancies due to differences in interpretation are resolved by the EPA Database Manager. Errors caused by incomplete Coding Guideline documentation are identified and modifications are made to the document.
3. Upon completion of the review process, the data coordinator checks to make sure the original reviewer's entry printouts contains the correct data, notes completion date on the "ECOTOX 10% Tracking Form". The ECOTOX 10% Replicate Review forms are filed with the double review entry printouts in the ECOTOX Reviewed files.

### 3. General Coding Information

#### Overview

ECOTOX is comprised of two databases - AQUIRE and TERRETOX. These databases were developed independently but are merging together under the ECOTOX framework. Across the two databases, the common data elements for each test contained in ECOTOX are grouped by chemical, organism, exposure conditions, and effects. Test chemical parameters describe the toxicant and any associated carrier; the CAS registry number; and the grade, purity and/or composition. The test organism parameters include the Scientific name, a species number and lifestage, source, and/or characteristics of the organism. The test conditions identify the test location; exposure type, time, and conditions; and any control parameters. Effect and endpoint parameters consist of codes to define lethal, sublethal, or residue effects and/or endpoints. The corresponding chemical concentration or dose is reported for both exposure and observation concentrations, if reported. Available data are extracted from the text, tables, and graphs of each publication.

Based on the information coded for the preceding categories, a documentation code is calculated for each piece of data in ECOTOX. The documentation code provides an index of the completeness of methods documentation and results presentation in the original publication.

#### Method for Prioritizing Studies for Inclusion in the ECOTOX Database

- For published Criteria Documents (Final and Draft Final), all data that appear in the tables will be encoded into the ECOTOX database.
- For the TRV project, all papers identified appropriate for use in deriving the TRV (Scores >65), and having MOR, REP, or GRO data will be coded into ECOTOX, without exception.
- Data maintenance reported by users or EPA, will be addressed in a timely manner, without exception.
- For criteria chemicals, where the Office of Water has only published the reference list, for all OPP-ESA papers, and for all general ECOTOX coding, the following papers will be prioritized for coding:
  - Study must be published as either a journal article, thesis or numbered government report. Conference Proceedings, extended abstracts, and unpublished materials will be marked as such and removed from the coding queue.
  - Studies must be published in English. Non-English publications will be marked as such and removed from the coding queue.
    - + Papers must report the use of a valid control. Studies that do not discuss the use of controls, or where the authors specifically state that a control was not used (ECOTOX codes NR and Z) will be marked as lacking a control (skim “No control” in the Unify skim module) and will be removed from the coding queue.
    - + The authors must report either a calculated endpoint, as defined by the ECOTOX coding guidelines, or presents statistically analyzed results. Papers presenting only qualitative data will be marked as such (“No endpoint, No stats” as noted during the skim processing and removed from the coding queue.
    - + Endpoint and/or statistical results must be summarized in either the text or tables. If results are only presented graphically or in figures, and specific concentrations (not ranged or approximate values) associated with the endpoint/statistical results are not presented, the paper is marked as having only graphed/figure data (skim “Graphed data only” in the Unify skim module), and removed from the coding queue.

The following sections are designed as an overview of the guidelines for reviewers. The information presented in this section identifies the common and unique attributes of each database. Each section heading corresponds to a data element (if the data element is unique to one or two of the databases, this is noted following the section heading). Any exceptions from these guidelines must be authorized by the EPA Database Manager and subsequently documented in these guidelines.

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## Coding Practices

This section provides an overview of the general coding practices used for the ECOTOX database. These practices have been devised to ensure accuracy and consistency in transcribing data from the original publication to the final data file.

- Each reported test exposure (or in some cases each unique endpoint) requires a separate Test entry into the Toxicity database.
- When endpoints, effect or exposure concentrations/doses, control data and exposure times reported in graphic format are coded (additional data was reported in text and/or table) the following are used to represent the data: Data extracted from graphs are presented as range or <, > values, unless an exact value is clearly presented. The values taken from the graph must be ranged using the author's X- and Y- axis values. Do not interpolate values that lie between axis values. A result remark is added to the observed response field denoting that the data were extracted from a graph. If the graph contains the only reference to a control, the availability of such data is noted in a corresponding REMARK, i.e., "control data graphed" is noted in the Control Remark data field. Data extracted from a graph must be accompanied by a comment in the REMARK field "from graph" in the corresponding remark data field. However, it is acceptable to estimate values from the graph for Exposure Durations (see Exposure Duration (ETIME) for further details). When there is a discrepancy between data presented in the text or table and data presented in a graph, the paper is to be forwarded to the EPA Database Manager for a final determination of which data point will be included in the database.
- To ensure completeness and accuracy, if information is unavailable for a coding field, the field must still be completed using either NR (not reported) or occasionally, NA (not applicable).
- To ensure accuracy in entering data values, all numbers between zero and one should be reported with a zero preceding the decimal point (e.g., 0.5 not .5). Periods are only used to represent a decimal point, never an abbreviation.
- To ensure consistency as well as accuracy, report the significant figures as the author reports them. Do not add or round off numbers. Report only the actual values, do not code variance information (e.g. +/-).
- When entering numbers do not use commas. They can be mistaken for decimal points or numbers.
- Use "per" or a colon (:) instead of a slash (/) to designate ratios. Reserve the slash for designating remarks or units.
- The REMARK field is a text field which contains additional information about a coding field. The REMARK field is used when the information necessary for coding a field does not fit in the space provided. A complete list of remark identifiers is documented in the appendices for each of the databases.
- When making a remark, use the appropriate codes from the ECOTOX code list. If a code does not exist for a certain chemical, enzyme, device, technique, etc., do not use the author's abbreviation. Write out the full name of the term and submit to the ECOTOX staff to determine if a new code should be created.
- When a reviewer has completed the review of a paper, they must write their last name preceded with "R=" on the bottom center of the paper.

- References that have data reported in the EPA Water Quality Criteria Documents (WQC) are coded into ECOTOX if it meets the ECOTOX criteria. If the data that is found in the WQC table does not meet the ECOTOX criteria, the data is not coded. However, the publication PDF is retained and a note is made in the bibliographic file stating that the paper was used in the WQC document, but the data was rejected and was not applicable to ECOTOX.
- If the author provides data from another publication, the reviewer should search the Unify reference module to determine if this reference has been previously identified as applicable to ECOTOX. If the paper has not been identified, the reviewer should have the paper ordered by the literature acquisition staff. If the cited data is from a publication which is within the ECOREF file, the ECOREF number should be noted on the original paper. Data should only be coded from the original publication to avoid duplicate data within the database.
- If publication is missing pages, pages are cut off or illegible, determine information is critical for encoding. Print out copy of the reference citation and write in the Order Notes field, a brief description regarding the incomplete status of the publication. For missing data that is critical to encoding the data, denote "critical for coding", and person to forward to, when updated information is received. If data are not critical, write "not critical for coding". For non-critical incomplete publications, the information will be updated, but reordering publication will be a lower priority.

For supplementary data published by journal and not printed, only attempt to locate the supplementary data yourself when needed for coding or are unsure if needed. If supplementary pages found AND verified critical for coding, print pages and place sticky note on front of paper "Add to PDF ...". Publication will be returned to EPA to ensure the printed copy will be added to the publication PDF. If the supplementary data are not critical, do not print, but make a notation for EPA to add the non-critical supplementary data to the PDF. If unable to locate the supplementary data and determined it is probably critical to encoding, print out citation forward to appropriate staff to reorder the publication with the supplementary data.

- All information from a paper is abstracted using the author's terminology and numeric values. Exceptions to this include the expansion of exponential numbers and when the author's "words" match the standard definition effects and endpoints. If an endpoint is "interpreted" by an abstractor, it is noted by an 'R' in the ASSIGNED P/R data field.
- Abstractors should note any comments about abstraction in the margins or on the tables/graphs of the original paper. This would include the Test ID Number for each unique test design, reason for data not being coded, LD50s outside of confidence intervals, errors between text and tables, or other anomalies. The abstractor encodes only the endpoint value and notes that the range was not coded in the original publication. Textual information is used over all other data, unless the value is noted in another section of the paper. Then, the most frequent value is encoded.
- Comments are used to better define or capture the researcher's intent. THESE ARE USED SPARINGLY. Comments are made in the associated data filed's

comment filed. For example, an ENDPOINT comment of median LC50 is located in ENDPOINT remarks data field as "median LC50".

- For all Comment data fields, reviewers should not use the percent or apostrophe symbols as these will cause errors to be thrown. The alternative is to omit apostrophes, and use the word for "percent" in the comment data fields.

## B. ECOTOX CODING GUIDELINES

### Quality Assurance Parameters

Note that all of the Quality assurance parameters are automatically updated by the Toxicity module and are found in the Paper Status data fields at the top right section of the entry screen.

Record History	(Modified)
Created by: csuomi	on 03/22/2013
Modified by: csuomi	on 04/29/2013
Citation QAed by: csuomi	on 03/29/2013

☐ Data Maintenance  
 Attachment

☒ Citation QAed    Unreviewed    Edit Paper Status

PAPER STATUS: Returned (En Route)    Checkout: 03/28/2013  
 DATA STATUS: REVIEWED - UNFY - COMPLETE    Checkin:  
 DUPLICATE STATUS: No duplicates    Last Returned: 04/29/2013

Recall    To Recall    File    Delete

TOTAL TESTS: 30    Last Reviewed: Brian Kinziger on 04/24/2013  
 TOTAL RESULTS: 54    Last RQAed: Amy Puglisi on 04/25/2013  
 TOTAL DOSE RESPONSES: 0    Last Maintenance QAed:

ECODEF#: 159926  
 Efficacy of Some Insecticides on Field Populations of Culex pipiens (Linnaeus) from Egypt by Emtithal, A.E.S., and A.E.B. Thanaa, 2012

Figure 1. Toxicity Module Quality Assurance Parameters

### QA Date/Initials

Once the person conducting the first Quality Assurance Check selects QA Complete, the Toxicity module marks the date of the QA check and the their name.

### Publication Reference Number, Author, Year

The Reference Number (Ref #) is the unique number which identifies a particular publication. This number, assigned by the data entry program, provides the link between the data entered and the original publication. It also displays the Author and Year.

### Total Tests

The total tests encoded for a publication are recorded by the Toxicity module. The total test number equals the total number of individual effect records that are coded for each publication.

Reviewer/Date

The reviewer's name and date of review is tracked by the Toxicity module.

Test Number (TESTID)

A new Test is added by searching on the Reference Number and selecting the review button. On the entry screen select Add Test. Select the test parameters that define the from the dropdown boxes that are populated from the Skim module.

Each unique test is assigned a unique test number (TEST ID) by the Toxicity module.. A unique test design may be characterized by a new test chemical, test species, test location, or exposure type. Additionally, there are experimental design parameters that will influence a test scenario sufficiently to warrant an independent Test TID. Such parameters include tests conducted at different test temperatures or conducted during different seasons. Some examples are found in Tables 1 & 2.

Table 1: A study is conducted with 2 different chemicals and the exposure for 2 species is started at 3 different lifestages. The Test IDs would be:

TEST ID	Unique Test Design
1	Benzene, Fish, Egg
2	Benzene, Fish, Juvenile
3	Benzene, Fish, Adult
4	Benzene, Invert, Egg
5	Benzene, Invert, Larvae
6	Benzene, Invert, Adult
7	Toluene, Fish, Egg
8	Toluene, Fish, Juvenile
9	Toluene, Fish, Adult
10	Toluene, Invert, Egg
11	Toluene, Invert, Larvae
12	Toluene, Invert, Adult

Table 2: A study is conducted with 1 chemical and the exposure for 1 species is conducted at 3 different temperatures. The Test IDs would be:



TEST ID	Unique Test Design	Temperature
1	Benzene, Fish, 15C	15 C
2	Benzene, Fish, 20C	20 C
3	Benzene, Fish, 25C	25 C

If appropriate, include information about the varying Experimental Design parameters in the Other Effect data field for each independent record coded. For example, if the temperatures varied as in table 2 directly above, TEMP EFCTS is coded in the Other Effect Data field.

### 1. Test Chemical Parameters (Chemical Information Tab)

ECOTOX is catalogued by the toxicant tested using the Chemical Abstracts Service (CAS) registry number. If a CAS registry number is not available through standard sources the toxicity data cannot be included in ECOTOX. Additional toxicants not included in ECOTOX are complex effluents, chemical mixtures and water/soil chemistry effects (e.g., pH) unless caused by a single toxicant (see Doses, Concentration and Media Characteristics for specifics on coding).

Chemical mixtures may be interpreted broadly. For example, a pesticide is a mixture of two active ingredients, each may have a separate CAS number. If formulation has a CAS number, the chemical reported for ECOTOX is the formulation. If the exposure is based on two metal compounds but the effect is based on one ion, e.g., copper sulfate and copper chloride and Cu is the toxicant, code copper as the test chemical and report the two compounds in chemical CHARACTERISTICS.

Chemical mixtures may be interpreted broadly. For example, a pesticide formulation is a mixture of two active ingredients, each ingredient may have a separate CAS number. If the formulation has a single unique CAS number, the chemical reported for ECOTOX is the formulation. If two different active ingredients are mixed together in a tank then applied as a toxicant, the data are not coded as this is a mixture.

If the exposure is based on two metal compounds but the effect is based on one ion, e.g., copper sulfate and copper chloride and Cu is the toxicant, code copper as the test chemical and report the two compounds in chemical CHARACTERISTICS.

If herbicides/fungicides are applied as standard weed/fungus control to both control and treatment groups prior to the application of the toxicant, the toxicant effects are coded and the application of the herbicide/fungicide is noted in the Experimental Design data field. If the chemicals are applied at the same time, this IS considered a mixture.

A similar example for lifestage studies occurs when the parental generation is exposed to one chemical prior to the gestational or post-gestational exposure of another chemical to the offspring produced, the parental data is coded until the second chemical exposure and the offspring data is coded only for the second chemical exposure. For the offspring records, it is noted in Experimental Design that the parents were exposed to a chemical prior to the gestational or post-gestational exposure.

Formulated or technical grade chemicals that report other chemicals as impurities are not considered mixtures.

Species exposed to more than one chemical during an exposure period are not necessarily considered mixtures. In some cases, the first chemical applied is used to treat a deficiency, disease, parasite, etc. to ensure more healthy test organisms or may be used to induce a biological response prior to a chemical exposure. When such a treatment is described by the authors, encode the beneficial or inducing treatment as an experimental design comment and encode the toxic effects of the other chemical(s) for ECOTOX.

For example, the authors state that “Grasses in all experimental sites were treated in the seedling stage of growth with bromoxynil for the control of broad-leafed weeds. No other herbicides were applied before the treatments with dicamba or propham”. Encode “Pretreated with bromoxynil in seedling stage for control of broad-leafed weeds” in the Experimental Design comment area on the Test tab of the Toxicity module. Encode the dicamba and propham toxicity data for ECOTOX.

A similar example for lifestage studies occurs when the parental generation is exposed to one chemical prior to the gestational or post-gestational exposure of another chemical to the offspring produced, the parental data is coded until the second chemical exposure and the offspring data is coded only for the second chemical exposure. For the offspring records, it is noted in Experimental Design that the parents were exposed to a chemical prior to the gestational or post-gestational exposure.

Another differentiation of a mixture is drawn when nutrients are added to an experimental set up. If the author states that a nutrient is added at a level that is needed for growth, ECOTOX does not consider this as a mixture. If, however, the author adds a series of nutrients and toxicants to test interactive effects, ECOTOX considers this a mixture. The following example illustrates how nutrients and toxicants are coded.

**A. Effect of copper on plant growth**

Nutrient Copper	20 ug/l	40 ug/l
0 ug/l	100%	120%
10 ug/l	100%	90%
20 ug/l	80%	80%
30 ug/l	60%	50%

**B. Effect of copper and nutrient on plant growth**

Nutrient Copper	0 ug/l	10 ug/l	20 ug/l	30 ug/l	40 ug/l
0 ug/l	100%	100%	100%	110%	120%
10 ug/l	100%	100%	100%	100%	90%
20 ug/l	95%	90%	80%	85%	80%
30 ug/l	60%	60%	60%	65%	50%



Nutrient	20 ug/l	40 ug/l
Copper		
40 ug/l	30%	20%

Nutrient	0 ug/l	10 ug/l	20 ug/l	30 ug/l	40 ug/l
Copper					
40 ug/l	20%	15%	30%	35%	30%

A. Author states that nutrients are added for growth. All results coded. Each nutrient level is coded as a separate test and the nutrient level is noted in an experimental design set-up comment in the EXP DESIGN field.

B. Author does not state that nutrient is added for growth. Two tests are coded, one for the nutrient tested alone and a second result for the copper tested alone. The shaded area is not coded. Mixture is noted in general remarks.

If the author does not document the basal level value, a determination may be possible for deficient, basal and toxic dose levels. The suggested guideline for making this determination would be interpreting dose response results for mortality, growth and reproduction to determine the deficient, basal (or basal range) and toxic levels. This could be accomplished by interpreting the trends for these effects. The deficient results would be excluded and basal level is coded as the control. If multiple basal values are reported, the most optimal dose for mortality, growth, reproduction would be considered the control value.

If the toxicant added does not produce a toxicity test result (i.e., only deficient and/or basal levels), then the paper would be rejected as nutrient study.

For *in situ* exposures where the exposure is by default an exposure to a chemical mixture; code residue effects or endpoints (BCF) only. No other effects or endpoints are strictly attributable to a single chemical in the same way as a residue concentration. Data for chemicals in the mixture with reported water concentrations and residue effects should be coded. The only situation in which a mixture exposure can be coded is when an *in situ* field study is conducted. The test organisms must be transplanted from a clean source and caged in the polluted source. The duration and concentration must be provided. When the author provides only the species age for the duration, it is unacceptable to code.

If the author reports that a precipitate was formed during the addition of the chemical or during the exposure period, code "precipitate formed" the EXP DESIGN field.

Effect responses from exposures to hormones (e.g. estradiol, testosterone) are included in the database. There are studies where the hormone is administered as a toxicant to observe the andro/estrogenic effects.

Studies involving carbon dioxide (CO<sub>2</sub>) or ozone (O<sub>3</sub>) as the toxicant are not coded into the ECOTOX database.

Lead shot papers are not encoded into ECOTOX. Authors often report the number of pellets fed to the animals, but not the actual dose of lead or concentration of lead/pellet.

There are also instances in which the lead pellet is actually a mixture of two metals (i.e. Lead and tin).

Nutrients such as phosphorus, nitrogen, potassium are coded for ECOTOX if the exposure system is dosed rather than an ambient exposure. For example, code phosphorus as an exposure chemical if, in the given paper, all of the following are true:

- The phosphorus was added to the ecosystem in a direct discrete manner, i.e., code "nylon mesh bags of  $\text{Ca}(\text{H}_2\text{PO}_4)_2$  placed in streams at beginning of test", do not code "system may have received added phosphorus in overland runoff due to fertilizers used in nearby agricultural operations". Aerial applications are acceptable if the other conditions are met.
- The concentration in the water should be measured, or at a minimum, the application rate should be available. Application rate may be calculated using the flow volume and the phosphorus-containing compound's dissolution rate.
- The effects of the phosphorus are tested on a biological test organism; water quality or chemical-fate only papers are not coded.

Effect responses from exposures to humic acid only are not coded for ECOTOX. Humic acids are any various complex organic acids obtained from humus which are insoluble in acids and organic solvents. However, tests that include an exposure with a toxicant and humic acid should not be interpreted as a mixture. The humic acid information should be coded into the humic acid field (see water chemistries section for specifics on coding) if the concentration is given and "Humic Acid Efcts" should be coded in the OTHER EFFECTS field.

Example: The toxicant is Copper and Humic acid is added at 10 mg/l.

Code the concentration of Humic acid in the Humic acid field as 10 mg/L .

Purpose	CAS Number	Name	Molecular Weight	Grade	Purity	Formulation	Radiolabel	Characteristics
Carrier	64175	Ethanol	46.069	NR	NR	NR	NR	NR

**Figure 2. Toxicity Module Chemical Information Tab**

### Chemical Name (TEST) and CAS Number (CAS NUMBER)

The chemical name as it is reported in the publication is selected from the entries entered into the Skim module. Common names will be populated to the Chemical Characteristics data display of ECOTOX. Reviewers should confirm the chemical entered at skimming is correct following the steps outlined below. If the name differs the reviewer must return to the Skim screen and update skimming prior to proceeding with the review.

If several names (e.g., trade names, synonyms) are used, note the other names and formula in the Chemical Characteristics data field.. In certain instances the author may

modify the test chemical by neutralizing or by adding a salt. The chemical that is introduced into the organism is what should be coded as the Test Chemical.

Example: Author reports purchasing hydrogen selenite CAS#7783008 which is neutralized with NaOH  
Author reports administering sodium selenite CAS#10102188 to organism.  
Code: Test Sodium selenite  
CAS Number 10102188

To ensure the correct chemical has been assigned during the skim process complete the following:

Check the Chemical module for the correct chemical name.

- If more than one CAS Number is retrieved, check the molecular formula
  - If molecular formula is presented, match the molecular formula to a formula in EcoChem for the given name. [Note: Check the abstract, methods, footnotes, table footnotes, and the Acknowledgments section for this information]
  - If molecular formula is not presented, do one of the following:
    - If a generic CAS number is available for the chemical, ensure that the generic compound has been skimmed to the paper
    - If a generic CAS number is not available, check to see if the metal has a common valence (see following table), e.g. copper chloride (the +2 valence is most common), assign the CAS Number for the most common valence for that chemical name.
    - For metal compounds that do not have a more common valence (see following table) and a generic CAS number is not available for the chemical name, assign the parent metal form, e.g. Arsenic if arsenate is the chemical reported by the authors.
    - For metal or nonmetal chemicals that do not fit these rules, refer the chemical to the Chemical verification staff for further investigation.
    - If the chemical in the paper is reported as a Trade name, check to see if an alternate name is presented in the paper to ensure correct chemical assignment. For example, the trade name Horizon is associated with three chemical active ingredients in the literature; Fenoxaprop-p-ethyl, Clodinafop-propargyl and Tebuconazole. Select the Horizon associated with the correct active ingredient. If no active ingredient is reported, forward to the chemical verification staff for additional verification.
    - If a hydrated form of a chemical is used in the paper, record the hydrated form as reported by the author in the Chemical Characteristics field. Also include the molecular formula if reported. However the CAS Number for the non-hydrated form is used for reporting the chemical and is entered as such in the skimming module (see the example below). However, if the paper reports a formulated product that is a hydrated compound (e.g. Borax which has a molecular formula of  $B_4Na_2O_7 \cdot 10H_2O$ ), the paper

should be skimmed/coded to the CAS number for the hydrated form rather than to the non-hydrated form as there is no cue for review staff that the compound is hydrated. If the paper were to report Sodium borate decahydrate instead (a synonym for Borax) then it would be coded to the non-hydrated form following the guidelines for hydrates mentioned above.

Example:

Chemical cited in paper - Copper Sulfate ( $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$ )

Code:

Chemical Copper sulfate ( $\text{CuSO}_4$ )

Chemical Characteristics  $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$

## Elements and Common Valences

(from Periodic Table of the Elements, Sargent-Welch, 1980 cat. # S-18806)

Element	Symbol	Common Valence	Element	Symbol	Common Valence
Actinium	Ac	3	Molybdenum	Mo	6
Aluminum	Al	3	Neodymium	Nd	3
Americium	Am	3	Neptunium	Np	5
Antimony	Sb	3	Nickel	Ni	2
Arsenic	As	3	Niobium	Nb	5
Astatine	At	-1	Nitrogen	N	3
Barium	Ba	2	Osmium	Os	4
Berkelium	Bk	3	Oxygen	O	-2
Beryllium	Be	2	Palladium	Pd	2
Bismuth	Bi	3	Phosphorus	P	5
Boron	Be	3	Platinum	Pt	4
Bromine	Br	-1	Plutonium	Pu	4
Cadmium	Cd	2	Polonium	Po	4
Calcium	Ca	2	Potassium	K	1
Californium	Cf	3	Praseodymium	Pr	3,4
Carbon	C	4	Promethium	Pm	3
Cerium	Ce	3	Protactinium	Pa	5
Cesium	Cs	1	Radium	Ra	2
Chlorine	Cl	-1	Rhenium	Re	7
Chromium	Cr	3	Rhodium	Rh	3
Cobalt	Co	2	Rubidium	Rb	1
Copper	Cu	2	Ruthenium	Ru	3,4
Curium	Cm	3	Samarium	Sm	3
Dysprosium	Dy	3	Scandium	Sc	3
Erbium	Er	3	Selenium	Se	-4
Europium	Eu	3	Silicon	Si	4
Fluorine	F	-1	Silver	Ag	1
Francium	Fr	1	Sodium	Na	1
Gadolinium	Gd	3	Strontium	Sr	2
Gallium	Ga	3	Sulfur	S	6
Germanium	Ge	4	Tantalum	Ta	5
Gold	Au	3	Technetium	Tc	7
Hafnium	Hf	4	Tellurium	Te	-4
Holmium	Ho	3	Terbium	Tb	3
Hydrogen	H	1	Thallium	Tl	1
Indium	In	3	Thorium	Th	4
Iodine	I	-1	Thulium	Tm	3
Iridium	Ir	4	Tin	Sn	4
Iron	Fe	3	Titanium	Ti	4
Lanthanum	La	3	Tungsten	W	6
Lead	Pb	2	Uranium	U	6
Lithium	Li	1	Vanadium	V	5
Lutetium	Lu	3	Ytterbium	Yb	3
Magnesium	Mg	2	Yttrium	Y	3
Manganese	Mn	2	Zinc	Zn	2
Mercury	Hg	2	Zirconium	Zr	4

### Chemical Grade (GRADE)

Enter relevant chemical grade information in the GRADE field. (refer to ECOTOX Appendix B).

A special note regarding: Technical grade vs technical formulation. Technical grade means the formulation contains small amounts of impurities. Technical formulation would mean the same thing; formulation includes small amounts of impurities. When this type of designation is made in a paper (e.g., technical carbaryl) the GRADE is coded as Technical Grade. Technical formulation is coded only if the author states "Technical formulation".

### Chemical Purity (PURITY)

Enter the numeric percentage information about the purity or active ingredient of the chemical in the PURITY data field (e.g., if the author reports 97% purity, 97 would be entered into this data field. PU for purity would be entered into the FORMULATION data field (see CHEMICAL FORMULATION).

### Chemical Formulation (FORM)

Enter the chemical formulation code for the chemical reported. If there is more than one formulation code enter the code most closely related to the chemical purity, and enter the rest in the CHARACTERISTICS field. (refer to ECOTOX Appendix C)

When authors report impurities in a formulated product (pesticide or organic compound), it will not be treated as a mixture, but rather as a discrete chemical.

The chemical formulation FORM is only coded when explicitly identified by the authors as the form (e.g., commercial) or when presented in a standard format (e.g., 25EC).

A special note regarding: Technical grade vs technical formulation. Technical grade means the formulation contains small amounts of impurities. Technical formulation would mean the same thing; formulation includes small amounts of impurities. When this type of designation is made in a paper (e.g., technical carbaryl) the GRADE is coded as Technical Grade. Technical formulation is coded only if the author states "Technical formulation".

### Chemical Comments (CHARACTERISTICS)

Enter relevant and specific chemical information, such as trade names, common names, isomers, or extra formulation codes, but do not code lot numbers or product numbers (usually taken from chemical catalogs).

**Note:** For NANOparticle papers, report particle size usually diameter, surface area and particle shape description (e.g. nano rods) in the Chemical Characteristics data field. If other information, e.g. Hydrodynamic size and Zeta potential is reported, "OTHER PARTICLE PARAMETERS REPORTED" is noted in the Chemical Characteristics.

### Radiolabel (RADIOLABEL)

If a radiolabeled chemical is tested, enter the isotope, according to the ECOTOX Appendix D codes, in the RADIOLABEL field. When the specific isotope is not reported, the RADIOLABEL remark field should be coded as "no isotope reported". When both

radiolabeled and unlabelled test chemicals are used in a test, report the radiolabel isotope and code "labeled and unlabelled" in CHARACTERISTICS.

If a test is conducted where a radiolabelled chemical is added to all experimental units and the non-radiolabelled chemical is added to all but one (authors may call this the control), and the authors do not report a clean control (nothing added), the test is then coded with Z control (no control reported). See ECOREF 9212 for an example.

### Solvent Chemical (S/V)

If a solvent carrier or Positive Control is used in the test, the solvent chemical fields are coded with the Solvent Chemical (S/V), GRADE, PURITY, FORM, concentration (in CHARACTERISTICS). The CAS numbers for common carriers are listed in ECOTOX Appendix A.

Occasionally two or more separate carriers or solvents are used. If the publication reports the ratio, include this information in the CHARACTERISTICS field. If the carrier or solvent is used for different test chemicals but the use is not specifically described in the publication, code "as needed" in the CHARACTERISTICS field.

If the publication reports data for multiple chemicals and reports the use of multiple solvents but does not report which solvent is used for which chemical, code all solvents for all chemicals and remark "1 of X solvents used" in the CHARACTERISTICS field.

Example: 10 pesticides tested, acetone and methanol used as solvents but it is not stated which solvent is used for each chemical, code:

2. S/V: ACETONE      CHARACTERISTICS: 1 of 2 solvents used

3. S/V: METHANOL      CHARACTERISTICS: 1 of 2 solvents used

If an author reports the concentration of the solvent used during the preparation of the stock solution and reports the concentration of solvent that the organism received, the concentration that the organism received is reported in the solvent CHARACTERISTICS field. If the concentration reported for the solvent in the control differs from the concentration dosed to the organism the concentration that the organism received is reported in the solvent CHARACTERISTICS field.

If a carrier was not used, report as NR. Buffers used to control the pH of the test are not coded. Acids or bases that are added to change the pH of a solution in order to enable a metal to stay in solution are not coded. Dietary feed content is not coded.

Occasionally, an author will present results data related to effect of the solvent on the test organism. If a test concentration and results are reported for the solvent and there is a separate clean water control, the solvent data should be coded.

If an author states that all solvent was evaporated prior to the study or if a column coating procedure is described, the solvent is assumed to not be incorporated into the study and should be coded as NR in the S/V field. Enter "'X' Solvent evaporated" in the test chemical characteristics, e.g. "acetone solvent evaporated".

For instances where a chemical carrier does not have a CAS number and it is determined by the chemical verification staff that it will never have a CAS number, the



solvent will be entered in the CHEMICAL COMMENTS field of the Chemical Name (TEST).

If the solvent does not have a CAS number enter the carrier/solvent name and characteristics in the Chemical Characteristics data field.

Example: The solvent used is Atlox at 0.05 ul/l. There is no CAS#

Entered into Chemical Comments: CARRIER ATLOX, 0.05 UL/L

If an author presents results in which the test organism is exposed to multiple chemicals, it is important to determine if the two chemicals constitute a mixture or if one of the chemicals is being used as a carrier or solvent. A carrier is defined as an agent (other than water) in which the test chemical is mixed to make it miscible with the dilution water before distribution to test chambers (Rand, 1995). If the other chemical is not a carrier, "mixture" would be noted in OTHER EFCT.

Water should not be coded as a solvent. A solvent is defined as an agent (other than water) in which the test chemical is mixed to make it miscible with dilution water before distribution to test chambers. Solvents or carriers are used in toxicity tests where the concentrations of the test chemical are extremely low and a very small amount of test material must be added to the test chambers. (Rand, 1995)

## 2. Test Organism Parameters (Species Information Tab)

Figure 3. Toxicity Module Species Information Tab

### Species (LATIN NAME/SPECIES NUMBER)

Select the species Scientific name as reported in the publication from the dropdown populated during the skim process. Refer to Species Procedures for additional information about the species data file and verification procedures. Field studies may report results for a target community (e.g. benthic macroinvertebrates) or for an entire enclosed ecosystem (e.g. system-level primary productivity or respiration). If a community of organisms was tested, be as specific as the author is about the species grouping.

For efficacy papers, the paper reports percent plants diseased (usually by fungus) or percent plants damaged (usually by insects or fungus) and the chemical applied is to reduce the amount of fungus or the number of insects, the effect is coded on the fungus or insects. These types of studies generally occur in the field.



**Examples:**

The author is testing peanut plants that are infected with a fungus. A fungicide is sprayed on the infected plants and the paper reports the “percent plants infected”. A population abundance (POP ABND) is coded for the Fungus and “measured as percent plants infected” is noted in Measurement Comments Field.

The author is conducting a field study where rice is sprayed with an insecticide to control insect pests. The paper reports the percent damaged plants and grain yield. A population abundance (POP ABND) is coded for the insects and “measured as percent damaged plants” is noted in Measurement Comments Field. The grain yield is coded as population biomass (POP BMAS) for the rice.

For terrestrial studies, decisions regarding the inclusion of species in ECOTOX are based on published terrestrial ecotoxicology standard methods and procedures documentation (eg., Menzer et al 1994; US EPA testing series; ASTM testing series). The focus for ECOTOX is to collect publications with data for soil invertebrate, plant species (agricultural and native), wildlife avian species (e.g. mallard, pheasant or bobwhite), wild mammalian species (e.g., meadow vole, deer mouse or mink), terrestrial lifestages of amphibians and reptiles, and beneficial invertebrate species (e.g., honey bee, leafcutter bee or alkali bee).

**Habitat (HAB)**

Select the habitat in which the organism is exposed; water for aquatic studies, soil for plant and organisms that live in soil or non-soil for organisms that do not live in soil.

The Habitat selection will enable different parameters for field data (Habitat Information Tab in the Test section) and Media Characteristics (Media Characteristics tab in the Result section).

**Organism Source (SOURCE)**

Select the source of the test organism (see Appendix E for codes). The source explicitly includes the strain of the organism, e.g. laboratory strain versus wild strain.

If the author(s) compare results based on the source of the organisms. "Organism source efcts" should also be noted in the OTHER EFCT field.

**Example:**

1st test: Commercial  
Source = COM  
OTH EFCT: Organism Source efcts

2nd Test: LABLaboratory  
Source = LAB  
OTH EFCT: Organism Source efcts

**Lifestage (LIFESTAGE)**

Select the specific lifestage for the test organism at the beginning of exposure, as reported in the paper (see Appendix F for lifestage codes). Record as 'NR' if the information is not reported in the publication. The following are examples of data coded in this field:

- a) The author reports adults are tested

Lifestage: AD

- b) The author reports exponential growth phase algae are tested

Lifestage: EX

Tests in which eggs are initially exposed, and the exposure continues through adulthood to the first generation etc, are represented as "EG" in LIFESTAGE and the stage of the organism is recorded in the Result Sample Unit field for the results reported.

Example: Exposed eggs resulting in mortality of fry

LIFESTAGE: EG    EFCT: MOR    MEASURE: MORT    Result Sample Unit:  
Fry

If data for multiple lifestages are presented along with statistical results, report results for each lifestage separately. If statistics are not presented, combine the results into one data record.

#### Age (AGE) and Age Unit (AGE UNIT)

Enter the age and age unit for each test organism at beginning of exposure, as reported in the paper (see Appendix I for associated time units). The age may be a development stage if no specific time is reported. For example: The author reports that 4<sup>th</sup> instar larvae were used in the study. The following is coded:

Lifestage: LV    Age: 4    Age Unit: ins

Record as 'NR' if the information is not reported in the publication.

#### Initial Weight (WTINT)

Report the weight and weight unit for each test organism at beginning of exposure, as reported in the paper.

#### Gender (Sex)

This field identifies the sex of the organism (male (M), female (F) or both (B) at each exposure level. The importance of this field becomes apparent where organisms of both sexes are exposed at a given treatment level, but the observations are conducted on either the male or female. In this instance, the SEX entry would be coded as B, with individual results reported for M and F in the Sample Number Unit field. Code 'NR' if not reported.

#### Organism Characteristics (ORGANISM CHARACTERISTICS)

Report any general information provided about the initial condition of the test organism that is not coded in the LIFESTAGE, INITIAL WEIGHT and/or AGE and AGE UNIT fields. This includes information for both the control and test organisms. Organism comments include information such as length, developmental stage, type of culture (eg., axenic) and/or initial cell concentration (e.g. 1 E + 3 cells/ml) to describe the organism being tested. Each piece of information is separated by a comma. The value and range, if reported, are recorded for each available parameter (e.g. 3(2-4) in). However, deviations are not coded (e.g. 33 +- 4 mm is coded as 33 mm). Record strains, hybrids

or taxonomic groupings, if reported. When reporting a cultivar, include 'cv.' before the name of the cultivar. Include 'var.' for variety in a similar manner. Common names and Taxonomy Author names are not coded in this field. List individual species Scientific names when 3 or fewer species are included within a grouping; when more than 3 individual species are included within a grouping, code as "# species."

Species = Plankton (#706)

ORG CHARACTERISTICS = *Daphnia magna*, *Daphnia pulex*, and *Bosmina sp*

ORG CHARACTERISTICS = 4 zooplankton species

Standard terms used for recording organism length include standard length (SL), (e.g. 3.1 cm SL), total length (TL), fork length (FL), carapace length (CL), carapace width (CW).

If a paper reports results using organisms from a polluted source AND organisms from a non-polluted source, only the non-polluted source test results are coded. No mention of 'non-polluted' needs to be presented in the ORGANISM CHARACTERISTICS field, but 'polluted organisms' should be entered into OTHER EFCT data field. If data are presented for organisms from a polluted source and no other concurrent data with organisms from a clean source are presented, the reviewer should code the test results, but enter 'polluted organism' in the ORGANISM CHARACTERISTICS data field.

If a paper reports that the organisms used for testing were diseased, "Diseased Organisms" is entered into the EXPERIMENTAL DESIGN field. If the study was conducted on diseased organisms compared to non-diseased organisms, the data from the non-diseased organism test is coded, but the data from the diseased organisms is not. In this case, "Diseased Organism Test" is entered into the OTHER EFCT field.

If a paper reports that the organisms used for the measurements were dead and it is unknown how long the organisms have been dead, do not code these data, but enter "dead organisms" in OTHER EFCT. An author may measure the growth or weight of a dead organism. The weights of dead animals can be biased, especially for juveniles, depending on the time between death and measurement. Also, temperature may effect the measurement after death (e.g. higher brooder temperatures may cause rapid desiccation which would result in weight changes).

If the study was conducted on dead organisms compared to living organisms, the data from the living organism is coded but the data from the dead organism test is not coded. In this case, dead organism is entered into the OTHER EFCT field.

If some of the organisms tested are fed differing amounts and/or some of the organisms are not fed during the study and the authors are comparing the data (e.g. 10 organisms fed, 10 organisms not fed and/or some organisms fed 10 pellets and some organisms fed 5 pellets), all data are coded, "Fed", "Not Fed" or food amount is coded in the EXP DESIGN field, and "Feeding Efcts" is coded in the OTHER EFCT field.

If an unverified species has be sent to the species verification staff and it was unable to be verified, the species verification staff will assign the next highest taxonomic level and the unverifiable name reported by the author is recorded in the ORGANISM CHARACTERISTICS field.

### 3. Test Information Parameters (Test Information Tab)

Figure 4. Toxicity Module Test Information Tab

#### Exposure and Study Durations

A toxicity test may range in duration from a pre-treatment period through the actual toxicant exposure and conclude with observations of the organisms post-exposure. Duration information is coded using the units reported in the publication (see Appendix I for valid units). Refer to Table 4 for a coding example. Exposure and study durations are reported with the Test Information. Observation Duration is reported with the Results Information.

Table 4. Example 17-day experimental period with 2-day pre-treatment, 5-day exposure, and 10-day observation. Note: Pre-treatment days are not included in the study duration.

	DURATION OF EXPERIMENT																
	Pre-Trt		Exposure					Observation									
Calendar Days	1	2	3	4	5	6	7	8	9	10	11*	12	13	14	15	16	17
Test Periods	1	2	1	2	3	4	5	1	2	3	4	5	6	7	8	9	10
Reported Days (Study Duration)	-2	-1	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15

**Note:** In test scenarios where incubation times are reported, i.e. enzyme fixation assays, be careful to report the toxicant exposure time *not* the assay incubation time.

### Study Duration

The study duration is the total time of the study *excluding* pre-treatment times. In the example in Table 4, the study duration is equal to 15D (5D exposure plus a 10D observation). In cases where the observation time is the only duration reported, it is assumed that the study duration is equivalent to the observation time. The study duration will be reported as 'NR' if no observation or study time is reported.

**Note:** for most field studies the exposure and study duration are identical because it is difficult to determine when the exposure ends. It is difficult to know when the application has completely dissipated in the environment. For lab studies the exposure and study duration may be different. This difference will be seen when there is a recovery period from exposure duration. For lab studies when the treatment is some type of injection or diet (intraperitoneally or by gavage), study duration and exposure duration are the same.

When study duration values are extrapolated from a graph the values can be approximated even when the mark does not appear on the graph. For example, if the value on the graph is in the middle of two marks namely 10 and 14 days, the STUDY DURATION can be coded as “12 d” with a remark that it is from the graph in the Study Duration comment data field.

### Exposure Duration

The exposure duration is a mandatory field for inclusion in the ECOTOX database. In cases where the observation time is the only duration reported, it is assumed that the exposure duration is equivalent to the observation time. If the exposure duration is not reported, the paper is rejected. The period of time recorded in the EXPOSURE DURATION data field is the time of actual exposure to the chemical. (See ECOTOX Appendix I for unit codes).

In some cases a biological time is used, such as an exposure time reported as “until hatch”, “growing season” or “after the nth egg has been laid”. Use the code from Appendix I that best describes the author's words in duration units data field. ‘NA’ will be coded as the duration mean value for all biological time durations (e.g. EXPOSURE MEAN: NA, EXPOSURE UNIT: HT; corresponds to an until hatch duration), unless a numeric value is substituted (e.g. EXPOSURE MEAN: 2, EXPOSURE DOSE: HV; corresponds to a 2 harvests duration). However, references to time such as “observed at end of the study period” are not coded; such papers are rejected as having no exposure duration.

For injection, diet, topical and environmental exposures where the actual exposure is dependent on biological and environmental conditions, the exposure time is recorded as equivalent to the study time. This assumption is made to ensure consistency in data representation; it is not necessarily a true reflection of the exposure time.

When exposure duration values are extrapolated from a graph the values can be approximated even when the mark does not appear on the graph. For example, if the value on the graph is in the middle of two marks namely 10 and 14 days, the EXPOSURE DURATION can be coded as “12 d” with a remark that it is from the graph in the Exposure Duration comment data field.

**NOTE:** For any single test record, only one duration unit can be used for this field. If authors report a combination of durations and they use different units for the same test, one of the values is converted to the other and summed (use only whole or decimal numbers). For example, the author reports 28 day of exposure followed by 8 hours of behavior testing (while still being exposed), the duration would be coded as 28.33 days. The 8 hours is divided by 24 hours in a day to get 0.33 days.

#### Application Frequency (APP FREQ)

Report the frequency of the dose application in the APPLICATION FREQUENCY data field. Refer to Appendix K for application frequency codes and units. If the exposure type is flow-through or continuous flow in lab studies, code "NR" as the frequency and "CON" as the unit in the APP FREQUENCY field. Examples of continual exposures for field data include artificial stream experimental systems and in situ exposures. If the exposure type for a lab study is static code "1" as the frequency and "X" as the unit in the APP FREQUENCY field. If an application frequency is not reported, record NR.

**Note:** The "X" in an application frequency unit represents the number that is written prior to the code.

#### Examples:

The author reports that the water was renewed every 48 hours. Code:

APP FREQ: 48 E X H

The author reports that the chemical was applied 3 times a month. Code:

APP FREQ: 3 X per MO

The author reports that the chemical was applied as a pulse dose of 3 hours every day. Code:

APP FREQ: 3 H per D

#### Control Type (CNTL)

The type of test control(s) used in the study is reported in this field. Control information for the reported effect may be presented in the text, in a graph, or in table format. ECOTOX reviewers do not make assessments whether the controls were satisfactory or insufficient (e.g., were replicates run, did control organisms die), but simply document whether the author(s) present information that a control was used. When author's state that controls were similar to treatment with the exception that no chemical was added, and within the same paragraph they describe using solvent in all treatments, a solvent control should be interpreted. (refer to ECOTOX Appendix M for control type codes and definitions.)

When statistical comparisons are presented for multiple controls (e.g., statistics in relation to a standard control and statistics in relation to a solvent control), enter all control types used in the study in the CONTROL TYPE field and note the appropriate control type used for the coded statistics in MEASUREMENT REMARK. (e.g., stats based on solvent control) The data related to the solvent control is coded preferentially over the standard control data and should be noted as such.



When multiple controls are reported enter all control into the Control data field.

### Concentration Type (CONC TYPE)

The three forms of toxicants evaluated in ECOTOX are organic compounds, metals and inorganic non-metals. Each form can be identified as a concentration type code using the single letter abbreviation.

Organic compounds are defined by the pesticidal terms, formulation (F) and active ingredient (A). Publications that do not specify the compound by the definition criteria for active ingredients are by default coded in the formulation (F) category.

Metals are defined by the concentration types, total (T), dissolved (D), and labile/free (L); while ammonia or hydrogen sulfide compounds may have total concentrations (T) and/or un-ionized (U) concentrations. Organometals are coded as total (T) concentrations.

If multiple representative concentrations of a metal or inorganic non-metal are reported in the reference, both concentrations are included in the same ECOTOX record; i.e., both total and un-ionized concentrations are reported in the concentration field. If the author reports the ammonia concentrations as based on  $\text{NH}_4\text{-N}$  or  $\text{NH}_3\text{-N}$ , code CONC TYPE as "T" and "U", respectively in the same record and code the specific ion information in the ION fields.

Concentration Type is also linked to the Chemical Analysis Method (METHOD) field (see discussion below on Active Ingredient).

### Concentration Types and Definitions

#### Organic:

**FORMULATION (F):** Way in which basic pesticide (toxicant) is prepared for practical use (Ware, 1978). Generally reserved for commercial preparation prior to actual use and does not include the final dilution (Insect-Pest Management and Control, 1971) (e.g.; Baythroid, 2,4-D). Also included in this category are organic compounds with no pesticidal activity (e.g.; PCB, dioxin).

**ACTIVE INGREDIENT (A):** Chemical substance in a product that is responsible for the pesticidal (toxic) effect (Ware, 1978). Reported as "A" when the author refers to the concentration as active ingredient, active principle, active substance (A.S.), acid equivalent or various grades of reagents (ie., Analytical, Reagent or Technical). When coding, a value in the publication may be reported as "Al kg/ha", "AE kg/ha", or "kg Al/ha"; in ECOTOX this type of value is reported as 'A' for CONC TYPE

If author reports both Formulation (F) and Active Ingredient (A) concentrations the Active Ingredient (A) concentrations are coded.

**Note:** Information reported in the PURITY field does not necessarily determine whether concentration is A or F. In addition to the description above, using "A" as the concentration type occurs in situations such as the following:

- 1) Author states concentration of pesticide as "Al".
- 2) Author states %Al (PURITY) and reports measured concentration.
- 3) Author states measured concentration of a pesticide.

#### Metal/Organometals/Inorganic metals:

**TOTAL (T):** The concentration of metals determined on an unfiltered sample after vigorous digestion, or the sum of the concentrations of metals in both dissolved and suspended fractions ( APHA et.al. 1992). Heavy metals and single elements (e.g. Na, Cl, Br) are coded as T.

**DISSOLVED (D):** Those constituents of an unacidified sample that pass through a 0.45 um membrane filter (e.g. soluble metal) (APHA et.al. 1992).

**LABILE (L):** The labile or free ion metal concentration determined by various analytical methods. When coding, the specific labile forms or complexes are not differentiated.

#### Inorganic non-metals:

Concentrations of ammonia and hydrogen sulfide are reported in the literature in either the total or unionized form. Code the form as specified by the author. Ammonia may be reported as a variety of different forms, eg.,  $\text{NH}_3$ ,  $\text{NH}_4^+$ ,  $\text{NH}_3\text{-N}$ ,  $\text{NH}_4\text{OH}$ , or  $\text{NH}_4\text{Cl}$ . (US EPA 1979) The author must state whether the form is **Total** or **Unionized**; **T** is the default for ammonia and hydrogen sulfide papers that do not state whether total or unionized concentrations are reported.

**TOTAL (T):** The dissociated, charged form of nitrogen or hydrogen related chemicals. This can take on numerous forms, e.g.; ammonium ( $\text{NH}_4^+$ ), nitrite ( $\text{NO}_2^-$ ), etc. (Rand and Petrocelli, 1985). **T** is the default for publications that do not state whether Total or Unionized concentrations are reported.

**UN-IONIZED (U):** The undissociated, uncharged form of ammonia or hydrogen sulfide. The ammonia molecule,  $\text{NH}_3$ , is the unionized form. (In aqueous solution, ammonia assumes an equilibrium between  $\text{NH}_3$  and  $\text{NH}_4^+$ .) The  $\text{NH}_3$  is the toxic entity of the ammonia compound (Rand and Petrocelli, 1985).

Some chemicals, e.g. aluminum compounds report many types of concentrations for aluminum: Total, Total Monomeric Inorganic, Reactive, Filterable, Dissolved, Labile, Non-Reactive, etc. In these cases, please only code Total, Dissolved and Labile when specifically reported by the authors. If other types, e.g. Total Monomeric Inorganic, Reactive, Filterable, Non-Reactive, etc, appear in the paper, add CHEMICAL FRACTIONATION in the Dose Comments field on the Test Information page for each concentration and in the Concentration Comments on the Results page for each result.

If they report the speciation of the compound, e.g.  $\text{AlOH}^+$ ,  $\text{Al}_2\text{O}_3^+$ , etc, add CHEMICAL SPECIATION to the Other Effects comments.

#### Study Type (STUDY TYPE)

For laboratory exposures, the study type is used to identify field simulation studies. For example, indoor mesocosm or microcosm studies should be noted as such in the STUDY TYPE field. If information about the study type is not reported, leave this field blank.

For field exposures record the study type as reported by the author in the STUDY TYPE field. If information about the study type is not reported, leave this field blank.

The valid codes for this field are:

ARTIFICIAL

CHANNEL

ENCLOSURE

LITTORAL

MESOCOSM

MICROCOSM



### Test Type (STYPE)

Report the type of test set up for the toxicity study as reported by the author (e.g. ACUTE, CHRONIC, ELS (Early Life Stage), FLC (Full Life Cycle), Generational). See ECOTOX Appendix HH for codes and definitions.

### Test Location (LAB, NR, FIELDN, FIELDA, FIELDU)

Enter the location or setting in which the experiment was conducted (see ECOTOX Appendix H). 'FieldX' selections will enable different parameters for field data (Habitat Information Tab in the Test section).

For example, a natural field study (FieldN) is an experiment conducted outdoors in a natural setting in which the test organisms are confined via an enclosure of some type (cage, fencing, plot lines) or sampled in the wild. Field exposures are exposed to uncontrolled variables such as weather. An important component for classification as natural is that the setting includes a bottom substrate as well as a community of representative organisms. Outdoor studies conducted in a simulated environment are coded as an artificial field study (Field A) study. Such studies include organisms isolated from their natural environment while still out of doors, e.g. earthen or concrete ponds without sediment or with only one representative species.

Laboratory tests are conducted under indoor controlled laboratory conditions (light and temperature regulated). If the location or setting cannot be determined from the publication code as Not Reported (NR).

### Test Method (TMETH)

Enter the name of the testing method used (or a modification of) for conducting the toxicity experiment (e.g. ASTM (American Society for Testing and Materials) or OECD (Organization for Economic Cooperation and Development). See ECOTOX Appendix II for the codes and definitions.

### Exposure Type (EXP TYP)

For the ECOTOX database, the term 'exposure' is used to refer to the mechanism by which the toxicant was applied. Organisms are typically exposed to toxicants through diet, injection, topical or environmental routes. On occasion, an exposure may be through multiple routes (e.g., such as topical and oral).

Some exposures could be coded a variety of ways. For example, exposure as an aerial spray to a field plot could be coded either as a spray application or as exposure through multiple routes, eg. topical (through skin) and diet (from consumption of exposed vegetation) for animals, or topical (through leaves) and environmental (root uptake) for plants. Within the ECOTOX database, this instance is coded as a spray application. Multiple exposure route coding is applicable when the organism is exposed through two *independent* applications, for example, a contaminated diet *and* topical application for animals or contaminated soil *and* leaf spray for plants. In this scenario, 'MU' would be

entered into the EXPOSURE TYPE data field and a remark ("FD and TP"/ or "PR and FS") would be noted in the Exposure Type comments section.

When coding, report the specific exposure type, e. g., for an intercutaneous injection, code as IC (intercutaneous). If an exposure type is not reported, code as Not Reported (NR). Refer to Appendix J for exposure type codes. If a species is implanted with a chemical capsule or time release pellet, code as the closest injecting type and note "Implant" in the Exposure Type notes field.

ECOTOX does not include in vitro assays [i.e. an experimental trial, involving biological matter, which is exposed to a toxicant in an artificial apparatus rather than within a living organism] in the database. Studies in which the living organism is exposed as a whole, but an effect on an internal process is examined outside the body after the exposure, are coded (e.g. enzyme functions). The database contains some studies using excised organs and cell cultures from plants, however these types of studies are not actively coded at this time. Future coding of these studies is under discussion.

ECOTOX does not include 'inhalation' studies or intratracheal instillation studies (placing chemical directly into lungs). These types of studies are rejected by using the keyword inhale.

### Media Type

Enter the type of exposure media, (e.g., natural or artificial soil, fresh or saline water), filter paper, etc.), in the EXPOSURE MEDIA data field using codes presented in Appendix L.

For terrestrial studies, if an aqueous exposure is conducted in pore water from a specific soil, report the soil parameters in the soil characteristics fields (pH, CEC, OM, etc.). If the bottom of the experimental chamber is covered with sand and then topped with filter paper, an Experimental Design (EDES) remark should be made that sand was used in the chamber, but MEDIA should be FLT.

For aquatic studies, freshwater (FW) tests include 1) laboratory tests conducted in freshwater, reconstituted water, distilled water, or tap water or 2) field tests where the organism habitat is exclusively freshwater. If a salinity value of <4 0/00 is reported and the paper does not specify whether it is fresh or saltwater, it will be coded as a freshwater test.

Saltwater (SW) tests include 1) laboratory tests conducted in natural or artificial seawater, brackish water, or estuarine water or 2) field tests where the organism habitat is exclusively saline.

If a determination cannot be made regarding the use of either freshwater or saltwater, an NR (not reported) is recorded.

### Number of Doses (DNUM)

Enter the total number of exposure doses, including the controls, for each independent test design. If number of exposures is not reported, e.g. in a publication reporting only calculated endpoints such as LC50s, code the field as 'NR'. Do not include number of replicates in the total number of doses.

### 3a. Doses (DOSES)

Table within the Test Information Tab)

There are multiple data fields that comprise a Dose Record; Dose Number, Control Type, Concentration Type Dose and Unit Organism Number and Dose Comments.

To Enter New Doses, select the Add Dose Link.

#### Dose Number

Dose Number is auto generated by the Toxicity Module. Doses are entered by Control first then in order of lowest to highest. The Dose numbers can be edited, if needed by selecting the Reorder Dose selection.

#### Control Type

Enter the Control Type used as needed. Only Control that were entered above will appear in the dropdown.

If a test is conducted where a radiolabelled chemical is added to all experimental units and the non-radiolabelled chemical is added to all but one (authors may call this the control), and the authors do not report a clean control (nothing added), the test is then coded with Z control (no control reported). See ECOREF 9212 for an example.

#### Concentration Type

Concentration Type is generated by the Toxicity Module base on the entry made above. If multiple Concentration Types are present (based on entries above) multiple rows will appear for population.

#### Doses

Enter the concentrations and the units used for the toxicity test in the Mean, Min, and Max data fields

If a background concentration is reported for the chemical being applied, report the background value in the control dose in the DOSE field.

Occasionally an author will report a concentration as a % or fraction of an LC50 value; e.g., either the sublethal concentration used was "10% of the 96-h LC50" or "1/10, 1/15 and 1/20 of the LC50". Such concentrations may be recalculated and used as the concentration tested if the original LC50 concentration is provided in the publication. Flag the recalculation in the paper so that the calculation may be QAed and document the recalculation in the margin or on a blank page of the publication. Put a comma after the concentration unit and remark "Recalculated" in the Dose comment data field. When multiple dose/concentration units are reported (e.g., ppm and mg/kg body wt), code the unit that represents the more accurate dose the organism receives (e.g., mg/kg body wt is more accurate than ppm).

For terrestrial studies, if chemical concentrations (especially metals) are reported in terms of Total, Exchangeable, Water Soluble and Pore Water concentrations, Total is

the concentration selected for entry into the dose data field. The other concentrations are reported as remarks.

For studies where a single chemical concentration is monitored as a water or soil chemistry parameter (e.g. sodium chloride reported as conductivity, calcium chloride reported as hardness), code the water or soil chemistry parameter in both the dose and the water chemistry parameter data fields.

### Organism Number

Enter the Number of organisms that were exposed at each dose. Do not sum all replicate values.

### Experimental Design (EXP DESIGN)

This field is used to code additional study information. For field tests, report exposure system dimensions (e.g. pond or lake depth, cage or enclosure size), type of artificial substrate and physical or chemical water chemistry parameters.

EXP DESIGN: 3 ha polyethylene lined pond//

EXP DESIGN: 4 x 4 m cage

EXP DESIGN: sediment

EXP DESIGN: Instant Ocean<sup>®</sup>

EXP DESIGN: Sinking Cr water

For laboratory studies, information about media and test chambers is coded if one of the purposes of the study is to compare results observed under differing test conditions (e.g., pH, temp, humic acid, sediment) or if commercial media types (e.g. Instant Ocean<sup>®</sup>) or Different soil types (e.g. OECD) were used in the study. If one of the purposes of the study is to compare experimental effects (pH, temp, sex) in addition to toxicant effects, report the additional effects in the OTHER EFCT field. (Refer to ECOTOX Appendix V for a list of keywords)

Example: OTHER EFCT: pH efct

Information about the dilution water is provided if needed to distinguish one test scenario from another, e.g. natural waters from three different ponds, sites on a river, locations in a sea. Tests with differing dilution water are coded as separate lines of data; it is not acceptable to combine tests by effect or water chemistry variables across differing dilution water test scenarios.

If an organism is pre-exposed to another chemical and this is the only information that can be coded, the chemical that is associated with the observed effect is coded and "preexposure with X" is noted in the EXP DESIGN field.

On occasion, an author will note that organisms were sampled during the course of a study for analysis. "Sub-sampled" should be noted in the EXP DESIGN field.

Example: Author tests organisms for residue and mortality over a 30 day study. At day 5 and 10, several organisms are pulled from the

study for residue analysis. At the end of the test (30d) the mortality is reported. "Sub-sampled" should be coded in EXP. DESIGN field since these sub-sampled organisms may or may not be included in the final calculation for mortality.

When coding field exposure publications, additional related coding parameters that do not get coded in the STUDY TYPE, HABITAT CODE, and SUBSTRATE fields are added to the EXPERIMENTAL DESIGN field. When adding field exposure information to the EXP DESIGN field, make sure to precede all data added with the appropriate field name code.

Example: EXP DESIGN: suspended mesh bags

If the author reports that a precipitate was formed during the addition of the chemical or during the exposure period, code "precipitate formed" in the EXP DESIGN field.

In field crop studies, sometimes the author uses standard cropping practices and may apply additional chemicals to the plots for weed and insect control or fertilization. If the author states that these are added as standard practice, MAINTENANCE CHEMICALS USED is added to the EXP DESIGN field.

#### Other Effects (OTHER EFCT)

Comments regarding other toxicity tests or effects reported in the publication that do not meet ECOTOX minimum data requirements are coded in this field. A keyword list (see ECOTOX Appendix V) for common terms is used as a guideline to assist the reviewer. The effect or endpoint codes are used when appropriate. The reviewer should maintain a list of new keywords and periodically submit this list to the EPA Database Manager. Commas separate each distinct term.

OTHER EFCT: uptake, elimination

OTHER EFCT: toxicity symptoms, diet study

OTHER EFCT: mixture, effluent

If other chemicals are tested as a mixture with the test chemical, the keyword "mixture" is coded in the OTHER EFCT field.

When water chemistry effects (temperature, salinity, pH) are tested in conjunction with chemical toxicity, a Remark is coded in OTHER EFCT to reflect this type of interaction.

OTHER EFCT: salinity efcts

#### Additional Comments

The Additional Comments data field contains historical comments from both the aquatic and terrestrial databases. No entries are made in the Additional Comments data field. If records are completed for data maintenance, the comments in the Additional Comments data field will be moved to the corresponding remark data field.

#### **4. Field Test Parameters (Habitat tab)**

(invoked when Test Location of 'FieldX' selected)

**Figure 5. Toxicity Module Habitat Information Tab**

### Sub-habitat (HABITAT CODE)

Select the one-letter code based on the Cowardin system code (see ECOTOX Appendix X) is used to describe the habitat (eg., Lacustrine or Riverine).

### Habitat Comment

The descriptor field is used to record the author's description of the water body, e.g. brackish marsh, oligotrophic lake, plastic tub, polyethylene lined enclosure. If the author does not provide any information about the habitat, both fields are coded as NR (not reported).

### Geographic Code (ST/PR/COUNTRY)

This field will contain the state, province or country name of the test site along with the Geo code. If the test site is not reported, an "NR" is coded. (ECOTOX Appendix BB contains a listing of country, region, province names and associated Geo code.)

### Geographic Code Comment (LOCATION)

Water body, city, county or relevant site information is coded. (see ECOTOX Appendix AA for field location abbreviations.)

### Longitude/Latitude (LAT/LONG)

If reported by the author the latitude and longitude are recorded. The "~" sign replaces the "°" sign in data entry. If not reported, NR is recorded.

An example of a longitude/latitude location (MED, Duluth, MN) is listed below:

Latitude: 46~50'51" N

Longitude: 92~11'12" W

An example of a ranged longitude/latitude location is listed below:

Latitude: 52~30'-53~30' N

Longitude: 107~30'-106~30' W

### Substrate (SUBSTRATE)

The bottom substrate is recorded as a two letter code by using the SUBSTRATE codes listed in ECOTOX Appendix Y. If there are no applicable codes, record as the author states in the literature. If a substrate is not reported, NR is recorded. A mixture of sediment types is coded as "MX" and should also include text for the most prevalent soil type(s) in the mixture.

Differentiate between organic and mineral soil/sediment by recording O for organic (leaves, detritus, debris) and M for mineral. Report % organic matter, if reported in literature.

#### Soil Type (Substrate Remark)

Report the scientific name of the natural soil or commercial name of the artificial soil used in the study. If the scientific name is not included report the type of soil using the author's terminology, e.g., forest soil, sandy loam soil, arboreal coniferous soil.

#### Application Type (AP TYPE)

This code will contain the method of application of the chemical. Application type codes are located in ECOTOX Appendix J.6.

For instances where the reviewer is unsure whether the chemical was applied directly to the water body by pumping, pouring, metering, etc., "DA" (Direct Application) will be coded.

#### Application Date (AP DATE)

The application date is the time of initial exposure. The format is mm-dd-yyyy, e.g. 12-01-1993. If one or more parts of the date is not reported, code the letter of the date that is missing (e.g. 12-dd-1993, mm-dd-1993, 06-15-yyyy). If more than one initial date is reported (e.g. more than one pond exposed), record the dates as a REMARK. If one pond is exposed multiple times, only report the first application date and note #x in frequency. If the application date is not reported, NR is recorded.

#### Application Season (AP SEASON)

This field is used **ONLY** if no application date is given by the author but the author does specify a season. This field contains the season of initial application of the chemical. A list of application seasons with dates and ECOTOX codes is presented below:

Code	Season
WI	Jan-March
SP	April-June
SU	July-Sept
AU	Oct-Dec
NR	Not Reported

#### Chemical Half-life (HALF-LIFE)

Record the specific chemical half-life in water as measured by the author. If the half-life is referenced from another paper, code this field as NR. If information about the half-life is not reported, record NR.

Example: HALF-LIFE: 2d

#### Application Rate (AP RATE)(Water Studies Only)

For Field tests:

This field contains the application rate value and the units that the author reports. If an application rate is not reported by the author, record as NR. If an exposure



concentration is not reported by author, the application rate must be reported. (See ECOTOX Appendix N for application units)

For Lab tests:

If an author reports an application rate, the information is coded in the following format: in the Dose Remark data field "Ap Rate/mean(min-max) unit/"

### Water Depth (DEPTH) (Water Studies Only)

Water depth value and unit are coded for the study site, as reported by the author. The software will convert the depth to a metric unit. "NR" is coded in the DEPTH field if the author does not report the water depth at the study site. If the author only reports the water depth of the entire system or the depth at which experimental units (i.e., cages) are suspended, "NR" is coded, and depth information is included in the EXP DESIGN field. (See ECOTOX Appendix Z for valid unit codes)

## 5. Result Parameters (Result Information tab)

Reference: **Skim** ECOREF#: 159926  
*Efficacy of Some Insecticides on Field Populations of Culex pipiens (Linnaeus) from Egypt by Emthath, A.E.S., and A.E.B. Thanaa, 2012*  
 Warnings: 1

ID	Chemical	Habitat	Species	Age	Lifestage	Study Type	Exposure Type	Test Location	#Results	Create Date	Modified Date	Collapse
2052459	101463698 - Cascade	Water	210 - Culex pipiens	3-4 hrs	LV	NR	S	LAB	2	04/24/2013	04/24/2013	

ID	Sample Number	Trend	Endpoint	Effect	Measurement (s)	Response Site (s)	Significance	Observed Duration	Concentration	Page Number	Reviewer Number	Modified Date	Collapse
2087923		INC	LC50	MOR	MORT	NR	NA	24 h	F 0.054 (0.044 - 0.063) ppm	65	1	04/24/2013	
2087925		INC	LC90	MOR	MORT	NR	NA	24 h	F 18.320 (15.83 - 20.80) ppm	65	2	04/24/2013	

**Result Information**

**Media Characteristic (Water)**

**Dose Response**

Sample size:

Endpoint:

Response Sites:

Projects:

Concentrations:

Significance:

Chemical Analysis Method:

Species Final Weight:

Percent Lipid:

Result Remarks:

Observed Duration:

Effect:

Effect Percent:

Significance Level:

Endpoint Assigned:

Intake Rate:

Dry Wet:

Steady State:

Lite Eval QA Comments:

Page Number:

Reviewer Number:

### Sample Number and Units (SAMPN)

The sample number reflects the sample size (e.g., 10 embryos) that the observation or result value is based on at each exposure level. For endpoints based on calculations (e.g. LD50, EC50 etc.) rather than individual dose measurements, the sample number will be coded as 'NR'. Code 'NR' if no information about the observed sample has been reported.



Sample units correspond to the sample number; i.e., the unit on which the measurement or endpoint is based (see Appendix Q for applicable codes). Code 'NR' if the sample unit is not reported.

**EXAMPLE:** For a sample size of 190 eggs, the sample unit is eggs (EG); therefore, if the effect measurement is HTCH, and the observation response value is 90%, then 90% of 190 eggs hatched.

**Note:** For generational studies and measurements based on the progeny, note F1, F2, etc. in the sample units field.

**Note:** If a sample number is not provided, but a "unit" is, always enter the unit in the sample units field.

**Note:** A FieldN test scenario involves exposing plots or sample areas, in addition to specific test organisms. Usually the number of exposed organisms is unknown. The number of plots or sample areas is coded as '#/EU' (the number of experimental units)

### Observed Duration (OBS TIME)

Observed duration is coded using the units reported in the publication (See Appendix I for Unit Codes). Time information may be extracted from a figure. When observed duration values are extrapolated from a graph the values can be approximated even when the mark does not appear on the graph. For example, if the value on the graph is in the middle of two marks namely 10 and 14 days, the OBS TIME can be coded as "12 d" with a remark that it is from the graph: Observation Duration Comment data field "from graph".

For a fluctuating or intermittent dosing (P) experiment, the total test time is recorded in the OBS TIME field with the exposure times and intervals between dosages reported in the APPLICATION FREQUENCY field.

Example: The author reports that the organisms were dosed with three pulses that were 45 minutes each in a 24 hour period. The test was run for 48 hours.

OBS TIME:48 H APP FREQ: 3 UNIT: X, 45 MI for 24 H

When an observation duration is not directly linked to a response, the observed duration is reported as the full range of time, e.g. "during a 10 week period" is coded as "0-10 wk" or if the observed response is for a portion of the exposure duration, ie., from day 2 through 10 wk, then code as 2-70 d.

For delayed effects, report the duration of exposure to the toxicant only in the Exposure Duration field. The observation time is recorded but will be greater than the Exposure duration. (See ECOTOX Appendix I for valid duration units)

If the duration is referenced from a WQC (Water Quality Criteria) document, code this as reported in the WQC document, and note that the duration is based on a comment in the paper and how it was coded in the WQC document.

For generational studies, report results only for lifestages that are directly exposed to the toxicant.

Example 1: Parent exposed - Offspring in clean water.  
Code effects on parents and code "generational study" in OTHER EFCT.

Example 2: Parents exposed - Offspring exposed  
Code the effects on both parents and offspring. Code the initial lifestage of organisms tested in the ORGANISM CHARACTERISTICS field and code the

lifestage of the organism being measured in the Sample Number and Unit field (SAMPN). The observed duration for the offspring is coded at the time the parents were exposed plus the time the offspring were exposed.

When coding the endpoints LTxx or ETxx, which are based on the time it takes to get a XX% response, code the associated effect response time and not the total exposure time in the observed duration field.

Example 1: A test with algae is run for 4 hours, but it takes 41 minutes for the "T1/2" duration which is the time of PSII half-inactivation. The author reported T1/2 endpoint is similar to an ET50 endpoint, therefore a reviewer assigned ET50 is coded.

Code: Assigned Endpoint: R      Endpoint: ET50      Effect: PHY      Measurement: PSII  
Observed Duration: 41 mi

Example 2: A 96 hour test is run with fathead minnows and reports mortality. The author reports the LT50 value at 10 ug/l, 20 ug/l and 30 ug/l as 3.5 days, 2 day and 1 day, respectively.

Code:  
Assigned Endpoint: P      Endpoint: LT50      Effect: MOR      Measurement: MORT  
Observed Duration: 3.5 d      Concentration: 10 ug/l  
Assigned Endpoint: P      Endpoint: LT50      Effect: MOR      Measurement: MORT  
Observed Duration: 2 d      Concentration: 20 ug/l  
Assigned Endpoint: P      Endpoint: LT50      Effect: MOR      Measurement: MORT  
Observed Duration: 1 d      Concentration: 30 ug/l

**Note:** For any single test record, only one duration unit can be used for this field. If authors report a combination of durations and they use different units for the same test, one of the values is converted to the other and summed (use only whole or decimal numbers). For example, the author reports 28 day of exposure followed by 8 hours of behavior testing, the duration would be coded as 28.33 days. The 8 hours is divided by 24 hours in a day to get 0.33 days.

**Note:** In test scenarios where incubation times are reported, e.g. enzyme fixation assays, be careful to report the toxicant exposure time *not* the assay incubation time.

**Note:** In test scenarios that involve generational studies, the observation duration times are reported from the time the parents were exposed. For example the parents were exposed for 10 months prior to mating, and the progeny was born 2 months later, the observation duration for both the adult REP PROG effect and for the juvenile DVP ABNL is 12 months. The exposure duration would be the same for both - 10 months. The only difference between the two effects is in the sample unit. The sample unit for the adult effect would be 'AD' and for the juveniles it would be 'F1'.

Toxicity test results for the ECOTOX database are represented by a combination of the ENDPOINT, TREND, EFFECT, RESP SITE, REMARKS, MEASURE, EFCT%, SIG, LEVEL, CONC and BCF fields. Toxicity test results for ECOTOX are primarily reported for observations taken during the chemical exposure; however, when results are reported *only* for the period of time after the exposure (moved to clean water), ie. recovery or delayed effects, this type of result is noted by using a "~" in conjunction with the endpoint/effect code, e.g. ~MOR for a delayed mortality effect.

ECOTOX codes data from papers in the following manner:

1. All author reported endpoints (from graphs, text and/or tables) will be encoded

2. NOEC/LOEC data points from statistically analyzed data at all measurements (i.e., author does not clearly identify these as NOEC/LOEC, but sig/nosig designations have been made), durations and response sites will be coded. These will be noted as "Reviewer Assigned"

**[Note:** Positive/beneficial effects (e.g. increase in growth over control, decrease in injury over control, increase in abundance over control) that are statistically significant are coded as NOEL/NOECs. A remark in the MEASUREMENT REMARK field must state that the effect was a statistically significant increase or decrease over control, which ever is applicable. For example, MEASUREMENT REMARK: statistically significant increase over control]

3. NR-ZERO and NR-LETH will be coded for aquatic and terrestrial studies following procedures outlined in the current ECOTOX coding guidelines
4. Data that are not statistically analyzed will be coded as a one line data marker, ranging all concentrations, durations and response sites per effect.
5. Data that are graphed and not statistically analyzed or data that is qualitative will be coded as a one line data marker, ranging all concentrations, durations and response sites per effect.
6. If the data encoded by a special project are not encompassed by the rules listed above (e.g. data from special projects that used the 10% change to assign endpoints), the data will be coded as follows:
  - For data reviewed by a special project prior to ECOTOX coding, not currently in the special projects database, the special project coder will note that a 10% endpoint was coded.
  - For data already in a Special project database, ECOTOX will extract a file of endpoints (noted by 10% text) in the endpoint comments (TRV) or statistic field (PCB) and examine the data encoded by the special project

For both of these cases, ECOTOX will add an entry (non-endpoint) to the database noting that a of 10% change occurred at the specific duration or concentration in the remarks field (e.g., 10% TRV).

Figure 1. Shows the hierarchy of coding tests in ECOTOX.

Endpoints always require a discrete line. For data not reporting an endpoint, at least one separate line is coded for each measurement from either a unique experimental design or within one design scenario for statistically defined data points. .

Food chain effects or endpoints are coded for organisms at the first level of exposure. Subsequent levels of exposure are not coded, but are noted in the OTHER EFECT: field, e.g. Other Effects: food chain study.

Endpoint, Effect, Measurement and Statistics sections have further description and examples.

The following sections provide a brief description for each of these fields, followed by guidance for coding information from the publication for each of the fields.

Endpoint (ENDPOINT)

For the purposes of ECOTOX, an endpoint is the quantification of an observed effect obtained through statistics or other means of calculation for the express purpose of comparing equivalent effects (e.g., LC50). ECOTOX Appendix T identifies and defines the ECOTOX endpoint codes. The endpoint field will be coded as NR if the author does not report or define an endpoint or there is no companion data point. All endpoints reported in the paper are coded.

Endpoint information is coded into ECOTOX if it is reported by the author, if the author's definition of the effect is equal to ECOTOX endpoint definitions, or if the data point is a companion endpoint to a LOEC, NOEC and/or MATC. "Companion endpoints" are endpoints assigned by the reviewer when the statistical results follow a clear concentration-response pattern and the author reports a NOEC, LOEC or MATC but fails to report the "companion endpoint". For example, when an author reports a NOEC and does not specifically define the lowest statistically significant effective concentration as a "LOEC", the data point is coded as a LOEC in ECOTOX by the reviewer. Similarly for reported LOECs without NOECs, NOEC/ LOECs without MATCs and MATCs without NOEC/LOECs.

**Note:** For terrestrial studies, and endpoints are reported for soil and pore water, code two separate endpoints for the soil and pore water endpoints. Add a Concentration remark "soil conc" or "pore water conc", respectively.

**Note:** Positive/beneficial effects (e.g. increase in growth over control, decrease in injury over control, increase in abundance over control) that are statistically significant are coded as NOEL/NOECs. A remark in the comments field must state that the effect was a statistically significant increase or decrease over control, which ever is applicable. For example, MEASUREMENT REMARK: statistically significant increase over control

If an author reports a NOEC and/or LOEC endpoint for one of the measurements in the text but does not specify the endpoints for the other measurements which show statistical data points within the same graph or table, the reviewer may extrapolate the SIG and NOSIG points for the other measurements as LOEC and NOEC using the R designation in the ASSIGNED ENDPT field. The reviewer may not extrapolate data from other tables or graphs. In studies where measurements are reported with statistical significance but the author does not report a NOEC and/or LOEC for any of the measurements, the reviewer does not code a NOEC and/or LOEC.

Example: The text contains the following statement: "The NOEC and LOEC for length were found to be 10 ug/l and 20 ug/l, respectively." A table reports weight data as well as weight and mortality data which is statistically analyzed.

Conc (ug/l)	Length (cm)	Weight (mg)	Mortality (%)	* Sig at p<0.05
Control	21	30	0	
5	22	30	5	
10	20	24*	5	
20	15*	20*	8	

Conc (ug/l)	Length (cm)	Weight (mg)	Mortality (%)	* Sig at p<0.05
30	13*	18*	25*	

Code:

Endpoint Assigned	Endpoint	Effect	Measurement	Conc	Signif	Level
P	NOEC	GRO	LGTH	10 ug/l	NOSIG	p<0.05
P	LOEC	GRO	LGTH	20 ug/l	SIG	p<0.05
R	NOEC	GRO	WGHT	5 ug/l	NOSIG	p<0.05
R	LOEC	GRO	WGHT	10 ug/l	SIG	p<0.05
R	NOEC	MOR	MORT	20 ug/l	NOSIG	p<0.05
R	LOEC	MOR	MORT	30 ug/l	SIG	p<0.05

On occasion, authors will report LC50 information in the methods section of their publication, without reporting any accompanying test procedure information. These test results are coded if enough information is provided by the authors to verify that the value(s) were not published elsewhere, and that the study meets all five minimal criteria for **acceptance**. The term "Methods LC50" is coded in the ENDPOINT REMARK.

If replicate tests resulting in a number of endpoints, (e.g. LC50s), are conducted, each LC50 must be reported on an independent line, even though the chemical, species, duration and effect are the same. Mean results are not coded if individual results are reported,

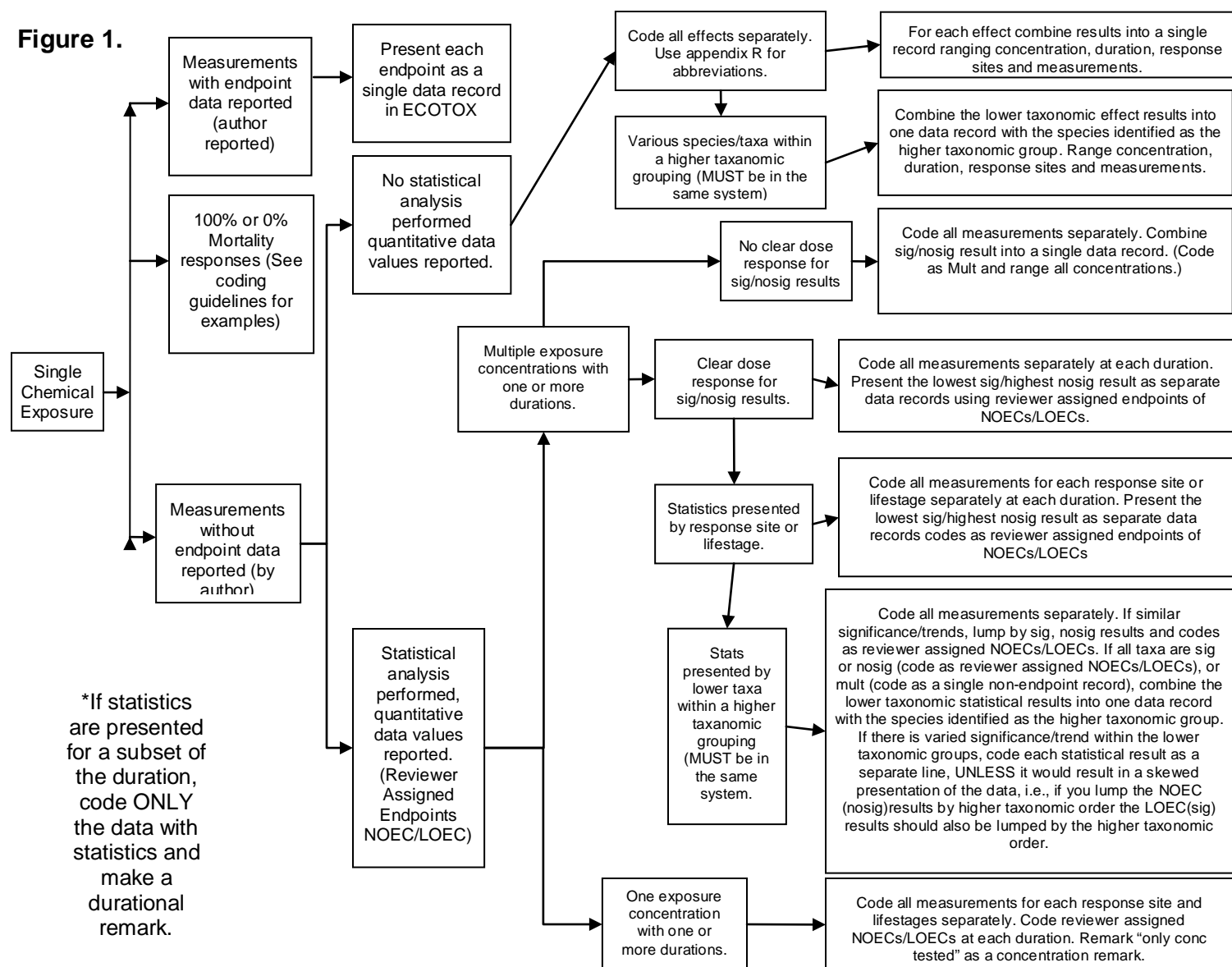
Example: Rep 1 - LC50 = 23 ug/L Code only Rep 1 and Rep 2.

Rep 2 - LC50 = 25 ug/L

Mean - LC50 = 24 ug/L

If a data set is evaluated using more than one statistical analysis all resulting endpoints are coded on separate lines (e.g. 2 LC50s for same data using probit and Spearman-Kärber will be coded as two separate data lines; report statistical method in ENDPOINT REMARK). Additionally, note "statistical comparison" in the OTHER EFCT field (see OTHER EFCT section for more information).

Figure 1.



Coding Hierarchy



## Endpoint Notes

If the author does not actually state that the value is an LD50 but states that “concentration x is the dose estimated to be lethal to 50% of the test organisms” and refers to statistical methods to estimate 50% lethality, the reviewer should code this as an LD50 endpoint because the author *defines* the LD50.

The ECOTOX database recognizes and codes “companion endpoints”; for ECOTOX such endpoints are defined as statistically significant endpoints that neighbor an author-defined NOEC or LOEC.

When a publication reports an author defined LOEC and NOEC for a non-monotonical response pattern (i.e., lower concentration significant and at least one higher concentration not significant); code the LOEC/NOEC reported by the authors, but note TREND as ‘CHG’ and code the SIG field as MULT to flag non-standard results.

In the ECOTOX database, the occurrence of no mortality (0%) or complete mortality (100%) is treated as an endpoint. The endpoints NR-LETH and NR-ZERO will always be coded for mortality effects of 100% mortality and 0% mortality, respectively. If an author reports 0% or 100% survival, ECOTOX will encode these data respectively, as follows:

Endpoint: NR-LETH Trend:INC Effect:MOR Measurement: MORT and Effect % 100

Endpoint: NR-ZERO Trend:NEF Effect:MOR Measurement: MORT and Effect % 0

If for a laboratory test exposure the authors report “all fish died”, code as NR-LETH and 100% mortality; however, for a field exposure, unless conducted in an enclosure of some type, it is difficult to assume that truly 100% of the fish are known to be dead, therefore the field exposure report of “all fish died” is coded POP, DEC, ABND, and EFCT% is not coded. The term “nil” is defined as “naught or nothing”, therefore, when used by an author, it will be assumed to mean 0% mortality and coded as NR-ZERO.

The 100% mortality data point at the lowest concentration/ shortest duration is coded. Similarly, the 0% mortality data point at the highest concentration/ longest duration is coded. In contrast to other endpoints, the additional mortality effects are coded along with the NR-LETH and NR-ZERO endpoint data. For example:



Mortality Table 1				
ug/L	24 H	48 H	72 H	96 H
1	0	0	0	<b>0 NR-ZERO</b>
2	5	17	30	35
3	25	40	65	90
4	<b>100 NR-LETH</b>	100	100	100

A) LC50s reported in publication, code

LC50s as reported

NR-LETH:4 ug/L at 24 hr

NR-ZERO:1 ug/L at 96 hr

B) LC50s not reported in publication, code

NR-LETH:4 ug/L at 24 hr

NR-ZERO:1 ug/L at 96 hr

MOR:2-3 ug/L at 24-96 hr EFCT%:5-90

Mortality Table 2				
ug/L	24 H	48 H	72 H	96 H
1	0	0	<b>0 NR-ZERO</b>	11
2	20	25	38	72
3	45	60	67	90
4	90	<b>100 NR-LETH</b>	100	100

A) LC50s reported in publication, code

LC50's as reported

NR-LETH:4 ug/L at 48 hr EFCT%: 100

NR-ZERO:1 ug/L at 72 hr EFCT%: 0

B) LC50s not reported in publication, code

NR-LETH:4 ug/L at 48 hr EFCT%: 100

NR-ZERO:1 ug/L at 72 hr EFCT%: 0

MOR1-4 ug/L at 24-96 hr EFCT%: 0-100

Mortality Table 3				
ug/L	24 H	48 H	72 H	96 H
1	<b>0 START</b>	0	7	13

Mortality Table 3				
ug/L	24 H	48 H	72 H	96 H
	RANGE			
2	0	28	45	60
3	38	44	67	100
4	38	60	100	<b>100 END RANGE</b>

A) LC50s reported in publication, code

LC50's as reported

B) LC50s not reported in publication, code

MOR1-4 ug/L at 24-96 hr EFCT% 0-100

Mortality Table 4				
ug/L	24 H	48 H	72 H	96 H
1	<b>0 START RANGE</b>	0	0	0
2	0	0	7	13
3	0	28	45	60
4	38	44	67	100
5	38	60	100	100
6	100	100	100	<b>100 END RANGE</b>

A) LC50s reported in publication, code

LC50's as reported

B) LC50s not reported in publication, code

MOR1-6 ug/l at 24-96 h, EFCT% 0-100

If the author reports an endpoint such as TLM, TL50, chronic value (ChV) or any terms with equivalent definitions that define endpoints such as those listed in ECOTOX Appendix T, the equivalent ECOTOX endpoint is coded in the ENDPOINT field.

If the author reports an endpoint that is a delayed exposure effect (~xxx), the delayed effect endpoint is coded if no similar exposure endpoint has been coded. A specific exception is gut clearance prior to tissue analysis; e.g., "after the exposure the organisms were placed in clean water for 10 hours to allow the organism to clear the stomach contents". This type of clearance is distinguished from depuration and is not coded as a delayed effect.

Trend (TREND)

The observed or measured response trend as compared to the control is coded when reported or graphically displayed.

When assigning a trend to a record, it should reflect the measurement which may or may not reflect the effect. For example, when authors report a decrease in survival; the effect is reported as MOR, and the trend is associated with the measurement; i.e., decrease in survival.

Example: EFCT: MOR TREND: DEC MEASURE: SURV

The trend for BCF, LCxx, LTxx is coded as "inc", except for the effect SVC (shell valve closure) which is coded as "dec". The trend for ECxx, NOEC, LOEC, and MATC will be either "inc", "dec", "chg" or NR depending on the results of the test. In instances when a trend is non-monotonical code "chg". The trend is noted as a two or three letter code:

<u>CODE:</u>	<u>TREND:</u>
INC	increase
DEC	decrease
NEF	no observed effect; e.g., when coding NR-ZERO the trend is NEF
CHG	no clear trend, results are variable (e.g. any combination of above trends listed)
NR	no trend reported <u>or</u> if no control response is reported then the trend is not able to be identified

Example: When a clear response, or lack thereof, is observed within an effect, it is coded as either INC, DEC, or NEF. The measurement used to evaluate the effect is reported in the MEASURE field, for example:

EFCT: GRO TREND: INC MEASURE: LGTH

When measurements do not report quantifiable data (see EFFECT MEASUREMENT section), data are combined into one record. If these data report multiple trends, code CHG in the TREND field and report the individual trends in MEASUREMENT REMARK field as in the following example:

Example: EFCT: HIS TREND: CHG MEASURE: EDMA,DISO,DEGN  
MEASUREMENT REMARK: inc EDMA, DISO, dec DEGN

Effect (EFFECT)

For ECOTOX database purposes, a toxicological effect is the observation or measurement of a response resulting from the action of a chemical stressor (e.g., mortality). The ECOTOX database internally categorizes all observed effects under at least one of eleven major effect group codes (Accumulation, Behavior, Biochemical, Cellular, Growth/Development, Lethal, Physiological, Population Community, Reproduction, Ecosystem and multiple groups). ECOTOX Appendix R describes the major groups and associated effect definitions for each three letter code. The major effect groups are not used by reviewers; their purpose is to provide database users the capability to search on broad groups of effects without specifying each individual effect. See Scientific Outreach Support for additional user support information.

The reported effect is interpreted to conform to the ECOTOX defined effects. If the effect is on the list of ECOTOX effects, use the ECOTOX effect code (see ECOTOX Appendix S). If the author's effect is not in Appendix S, but is similar to one already defined use the ECOTOX code which matches the definition and note the author's effect term in the MEASUREMENT REMARK field. If the author's effect appears to be a new effect code, discuss and forward to EPA Data Manager for approval.

Listed at the end of ECOTOX Appendix R there are two special effect code conventions used in ECOTOX. The first is NOC (No effect code) used only for ENDPOINTS reported by the author as ns, e.g. "mortality and growth" and no specific effect code can be assigned. This code is used *only* when such effects cannot be separated into or reported as individual effects. The NOC code is rarely used and when used must be verified by one or more fully trained reviewers.

The second effect code convention is ~XXX to indicate that the result reported was observed after the exposure period ended and the organisms are observed in clean water, i.e., a delayed response. Within a publication, delayed response data is reported only if exposure period observations are not available for the same effect or endpoint. When delayed response data accompanies exposure period observations, the delayed response data is not coded but is recorded in OTHER EFCT as "recovery".

**NOTE:** A specific exception is gut clearance prior to tissue analysis; e.g., "after the exposure the organisms were placed in clean water for 10 hours to allow the organism to clear the stomach contents". This type of clearance is distinguished from depuration and is not coded as a delayed effect.

Growth data reported on a part of an organism (i.e. weight or length of a specific organ/tissue/structure) is coded as MPH while growth data for the organism as a whole is coded as GRO. Roots and shoots of plants can regenerate and are considered a whole organism.

Occasionally, effects describing a parasite-host relationship are coded in ECOTOX. For example, the effect on the host is typically coded as an IMM effect with the measurement code PRNF. The effect on the parasite is typically coded as a POP effect with the measurement code ABND.

### Effect Hierarchy

- A. If the author has defined an Endpoint for an effect, report the Endpoint as outlined in the preceding ENDPOINT HIERARCHY.
- B. When only effects are reported in the publication, no endpoints, code the concurrent effects (results reported concurrent with exposure to chemical) according to the abbreviations in ECOTOX Appendix S. Code NR (not reported) in the ENDPOINT field.
  - i. If statistics are presented in a clear dose response, code the lowest significant effect as a LOEC and the highest nosig levels as a NOEC and appropriate p-values at all Durations, Measurements and Response Sites. Data must be monitonical at all durations to encode NOECs and LOECs.
  - ii. If statistics are presented and there is no clear dose response, code the ENDPOINT as NR, the significance as MULT and the appropriate value

for each Measurement and Response Site. The Durations and Concentrations are ranged.

- iii. If no statistics are used, or reported, combine the data by effect by coding a range for concentration and durations. all Measurements are encoded into the Measurement data field. Report as NR in the SIG and LEVEL fields. If these data report multiple trends, code CHG in the TREND field and report the individual trends in MEASUREMENT REMARK field as in the following example:

Example: EFCT: HIS TREND: CHG MEASURE:  
EDMA,DISO,DEGN  
MEASUREMENT REMARK: inc EDMA, DISO, dec DEGN

- C. When the only effects that are reported are those subsequent to exposure, report these as delayed effects, noted with a ~ preceding the three-letter effect code, e.g. ~MOR. Follow the procedures outlined in Steps B i, ii, iii for reporting delayed effects.

### Response Site (RESP SITE)

A response site code is used to identify specific organ and tissue effect sites for residue, biochemical, cellular and/or physiological effect measurements. Response sites are used for the effect codes ACC, BCM, ENZ, HRM, CEL, GEN, HIS, INJ, IMM, PHY, GRO, and MPH effects and associated endpoints. The response site is coded as NR for the effect codes MOR, AVO, BEH, FDB, DVP, ITX, POP, REP and PRS. The two or three letter response site codes are listed in ECOTOX Appendix U. It is acceptable to code a response site when tissues or organisms are pooled together for a measurement.

If a response site is not reported or not applicable, e.g. mortality, behavioral effects, code the site as Not Reported (NR).

If data for a number of tissues are presented along with statistical results report results for each tissue separately. If statistics are not presented, combine the results into one data record by entering each response site into the Response Site data field

### Combining Response Site

When the residue measured in one organ or tissue is further analyzed to indicate concentrations in cells or cellular fractions, a comment is placed in the Response REMARK field (e.g., "subcellular fraction" or "subcellular distribution").

If multiple tissues are being combined into one record, enter all of the the individual tissues/organs are coded in the Response Site data field. If the response site does not have a response site code request a new code be added. When the response site is not reported, the field is coded as NR.

### Measurement (MEASURE)

Generally, "measures" or "measurements" are variables used to aid in the interpretation of the degree of response to a toxicant by an organism. For example, measures of behavioral effects in ECOTOX include behavioral changes (BEH SWIM), changes in feeding activity (FDB FDNG), and stimulus avoidance (AVO STIM). ECOTOX Appendix

S lists the measurements currently used for each of the effects in the ECOTOX database.

If more than one organism is measured in either an artificial (pot, cage, aquaria, etc.) or natural population (field, water body, etc.), reviewing staff will code the effect and measurement as POP BMAS (population biomass). For instances where there is an individual biomass (one organism, artificial or natural), reviewing staff will code GRO BMAS (growth biomass).

Note: Growth data reported on a part of an organism (i.e. weight or length of a specific organ/tissue/structure) is coded as MPH while growth data for the organism as a whole is coded as GRO. Roots and shoots of plants can regenerate and are considered a whole organism.

Each measurement for which an endpoint is reported in the text, tables, or graphs receives a separate data line. However, measurements that are not statistically analyzed, or are discussed in the text and do not report any numeric values may be combined into one record. If several measurements are combined, code the General measurement code in the MEASURE field and list the separate measurements in the following format: MSMT/code,code,code//. See example 2 and 3 directly below.

For Gene Expression studies, authors will report measurements as either the genes that were expressed (e.g. Zgc:92628, Signal recognition particle 14 kDa, ATPase family – AAA domain containing 3A) or as a specific mRNA (e.g. CYPmRNA). Authors tend to use targeted techniques like Real-Time Quantative PCR or Protein Blotting, etc., and transcription processes like Microarray or Next Generation sequencing.

- ☐ In ECOTOX, if the author uses the targeted techniques, code each measurement separately. If the authors clearly identify a primer was used, note “Primer used” in the Measurement Comment data field.
- ☐ If the author uses transcription processes, code a maximum of one NOEC and/or one LOEC or one NR record for all of the expressed genes. Add a comment in the Measurement Comment data field noting the number of genes expressed. If the numbers of up and down regulated genes are reported add the specific comment ‘XXX genes upregulated and XXX downregulated.’ If the author clearly identifies GEO or CEB Accession numbers, note “Accession numbers reported” in the Measurement Comment data field.
- ☐ If the author uses both targeted and transcription processes in the paper, code the targeted data as noted above and report “Gene Transcription Analysis also reported” in the Other Effects data field.

Example 1: Length and weight reported in table statistically analyzed.

CONC	0	10 ug/l	20 ug/l	30 ug/l	40ug/L
Length	-	nosig	nosig	sig	sig
Weight	-	nosig	sig	sig	sig

Code: Endpoint: NOEC EFCT: GRO MEASURE: LGTH CONC: 20ug/L - nosig  
 Endpoint: LOEC EFCT: GRO MEASURE: LGTH CONC: 30ug/L - sig

Endpoint: NOEC EFCT: GRO MEASURE: WGHT CONC: 10ug/L - nosig  
 Endpoint: LOEC EFCT: GRO MEASURE: WGHT CONC: 20ug/L - sig

Example 2: A decrease in length and weight reported on graph which is not statistically analyzed but values are given.

CONC (from graph) 0 10 ug/l 20 ug/l 30ug/L  
 Code: EFCT: GRO TREND: DEC MEASURE: GGRO CONC: 10-30ug/L  
 MSMT/WGHT, LGTH//

Example 3: Histology reported in text. At concentrations of 10 ug/l , 20 ug/l and 40 ug/l, Reports that liver had edema, swelling, and hypertrophy.

Code: EFCT: HIS MEASURE: GHIS MSMT/EDMA, SWEL, HYPT//  
 CONC: 10-40 ug/l

More information on coding statistics is found in the statistics section of this SOP.

Many publications which report field data or laboratory microcosm studies present results for multiple species/taxonomic groups. The combining of results for species and taxonomic groups depends on whether statistics were applied to the data and whether a similar response is evident.

- If, within a higher taxonomic group (e.g., Algae), individual effects for several lower taxonomic groups are also presented (e.g., Bacillariophyta (diatoms), Chlorophycota (green algae), Pyrrophyphyta (dinoflagellates)) the data may be reported in a number of ways. Examples include:

- The measurements within each group are statistically analyzed and are similar overall, ie., INCreasing, DECreasing or CHanGing. Combine the results and code as:

Species: Algae ORG CHARAC: 3 orders EFFECT: POP MEASURE: ABND  
 TREND: INC SIG: SIG

- The measurements within each group are statistically analyzed and differ from each other. Code each result as a separate line:

Species: Bacillariophyta EFFECT: POP MEASURE: ABND TREND: INC SIG: SIG  
 Species: Chlorophycota EFFECT: POP MEASURE: ABND TREND: DEC SIG: SIG

- If no statistical analysis has been reported, the results from the lower taxonomic groups can be combined into a single record representing the next highest representative taxonomic group.

Species: Algae ORG CHARAC: 3 orders EFFECT: POP MEASURE: ABND  
 TREND: CHG SIG: NR

### Effect % (Effect Percent)

The Effect Percent field is used when the effect is reported as a percent change, e.g. percent of the total population or percent increase or decrease or if the percent response or raw data response is reported.

If the author reports the number dead (i.e., "5 of 20") do not recalculate as a percent.



Example 1:	80% mortality	EFCT: MOR MEASURE: MORT	TREND: INC	Effect Percent: 80
Example 2:	25% survival	EFCT: MOR MEASURE: SURV	TREND: DEC	Effect Percent: 25
Example 3:	5 of 20 died	EFCT: MOR MEASURE: MORT	TREND: INC	Effect Percent: NR
Example 4:	45% inc ATPase activity	Effect Percent: 45	EFCT: ENZ Measure: ATPA	TREND: INC
Example 5:	53.33% Hematocrit	Effect Percent: 55.33	EFCT: BCM Measure: HMCT	

If the percent effect is coded from a graph, code the percent values using a qualifier, ie. <, >, or ~, using only the graphical intervals reported on the graph. Code, from graph, in the Effect Percent notes field. If the percent effect is graphed and is not clear enough to extrapolate, code "graphed" in Effect Percent notes field. If the effect percent is not reported, the field is coded as NR.

If the percent effect is presented as "xx% of the control, Code the percent in the field and code: of control in the Effect Percent notes field.

Example: GRO WGHT Dec 20-30% of the control  
 EFCT: GRO MEASURE: WGHT Effect Percent: NR 20-30 TREND: DEC  
 Notes: of control

For NR-ZERO and NR-LETH endpoints, the Effect Percent field is coded as 0% and 100%, MORT respectively.

When no stats and effect % are presented at the highest concentration, the concentrations should be ranged and the Effect Percent should be presented as < the effect % from the paper.

### Combining Effect Percent

When data for an effect are combined because a statistical analysis was not applied and/or a clear dose response was not observed, and several percent effect values are presented, there are two different ways to report data.

If the author reports the effect measurement on a single parameter, the effect percent is reported as a range.

Example 1:	30-75% mortality	EFCT: MOR	MEASURE: MORT
	TREND: INC	Effect Percent: 30-75	
Example 2:	20-30% dec O2 consumption	EFCT: PHY	MEASURE: OXYG
	TREND: DEC	Effect Percent: 20-30	

### Projects (This data field is not encoded by reviewers)

The intent of this data field is to link results to an existing data set, i.e. CAD, TRV, EcoSSL, EFED, etc. This data field is NOT populated by reviewers, but rather by the person linking the data record to the existing data set or by the TOXICITY module when

delivering the EFED data. It is used by ASTER to include evaluated data into the ASTER profile report.

### 5a. Concentration Parameters

(Table within the Results tab)

#### Concentration Type (CONC TYPE)

Concentration Type is populated from the entry made on the Test Information tab.

#### Ionic Fraction (ION)

For ionizing substances (e.g., metals, ammonia), report the dose as the ion if the concentration presented by the authors is reported as based on the ionic form of the compound (e.g., organotin as Sn). Code the appropriate ionic symbol in the ION field (see ECOTOX Appendix O for Ion codes). If concentration is based on the total compound, code 'NR' in this field. For non-ionizing substances, code 'NR' in this field.

If the test chemical for a metal is reported as the elemental metal (i.e. mercury) code the ion (Hg) in the ION field.

#### Effect Concentration (CONC)

Report the effect concentration in the same units used by the author. If the author reports that the concentration is based on active ingredient or acid equivalent choose the unit preceded by AI or ae from the dropdown menu (e.g. AI mg/kg or ae mg/kg). Do not convert any units. (See ECOTOX Appendix N for a list of concentration units). The confidence interval, fiducial limits, or range is recorded when available. The media concentration is coded in this field, except for diet studies, where the concentration in the food is coded. If a test is run with two sources of chemical, such as diet and water, code the concentration of the diet in the CONC field, the EXP TYPE field as FD and code "water conc rpt" in the Concentration REMARK field. If the concentration reported from spiked sediment studies is the pore or overlying water concentration, record the concentration in the CONC field, and code "pore water from spiked sediment" or "overlying water from spiked sediment" in the ConcentrationREMARK field and ensure that the Exposure Type is 'L' (leachate).

Measured concentration data are selected over nominal concentration results, except when the presentation of the results is more complete with the nominal concentration data vs. the measured data (i.e., lumping records vs. separate test results).

When coding numbers do not use commas. They can be mistaken for decimal points or numbers.

Code the mean and range of a stated concentration unless there are multiple test results being combined into one test record. In this case the lowest minimum concentration and the highest maximum concentration will be used.

Example 1:      Endpoint reported as LC50 = 100 (50-150) ug/L  
                     Code: ENDPOINT: LC50 CONC: 100 (50-150) ug/L

Example 2:      Effect (reported at 1 concentration tested with replicates)  
                     Rep1 = 10 (9-13) ug/L

Rep2 = 11 (8-13) ug/L

Code: The lowest minimum value and the highest maximum value are coded.

Conc: 8-13 ug/L

Example 3: Multiple concentrations reported with no statistics. Combine concentrations into one record coding the lowest minimum value and highest maximum value.

Nominal Conc.Measured Conc.

1 ug/L1.5 (1-2) ug/L

2 ug/L2.3 (2-3) ug/L

3 ug/L3.1 (2.5-3.3) ug/L

Code: Conc: 1 -3.3 ug/L

If the reported mean value does not fall within the reported range, confidence limits or interval, code the mean value in the Effect Concentration field. In the Concentration Remark field, code: "CL (or CI) reported as XX-XX" or "range reported as XX-XX", where XX-XX is the reported range, confidence interval or limit.

Occasionally an author will report a concentration as a % or fraction of an LC50 value; e.g., either the sublethal concentration used was "10% of the 96-h LC50" or "1/10, 1/15 and 1/20 of the LC50". Such concentrations may be recalculated and used as the effect concentration if the original LC50 concentration is provided in the publication. Flag the recalculation in the paper so that the calculation may be QA'd and document the recalculation in the margin or on a blank page of the publication. Put a slash next to the concentration value and note in the Concentration REMARK field: "Recalculated".

When concentrations are taken from a graph, put a slash next to the concentration value and note in the Concentration REMARK field: "from graph".

All reported concentrations are coded and identified as to whether the concentration is based on the active ingredient or formulation, or as the total, un-ionized or dissolved concentration, are identified (see CONC TYPE).

In certain cases, the ECOTOX concentration is routinely reported as some form of the test chemical. For metal salts, the concentration is generally expressed as ug ion/L (e.g., Hg<sup>+</sup>). Be sure to code the ION field with appropriate ion.

An exponential number greater than +8 or smaller than -7 (e.g.,  $1 \times 10^8$ ; often reported as  $10^8$ ) is coded as E+n or E-n (e.g., 1 E+8). The concentration field is 10 characters long, therefore numbers less than or equal to +8 or -7 can be written out, eg.  $10^6$  is reported as 1000000.

When the concentration is reported as the metal (e.g., Sn), but the chemical tested is identified as an organometallic (tributyltin chloride ( $C_{12}H_{27}ClSn$ )): Record the full name of the chemical tested in the TEST field, enter "T" in the CONC TYPE, report the concentration in the CONC field, and identify in the ion that the concentration is based on the metal component and report in the ION field.

For studies where a single chemical concentration is monitored as a water or soil chemistry parameter (e.g. sodium chloride reported as conductivity, calcium chloride

reported as hardness), code the water or soil chemistry parameter in both the dose and the water chemistry parameter data fields.

If the author states that the control water was from a polluted source or was contaminated, 'contaminated controls' should be noted in the EXP DESIGN field. The concentration of chemical in the controls is not coded. If the author is reporting the chemical concentration of a control which is a background level, this information is not coded.

If the chemical used is an aged solution and the author is comparing the data to varying aged solutions (e.g. 2 d aged solution, 4 d aged solution and 6 d aged solution), all data are coded, "X d aged solution" is coded in the EXP DESIGN field, and "Aged Solution Efcts" is coded in the OTHER EFCT field.

For field data, the water concentration may be reported as NR, if the application rate is reported (see AP RATE field). However, the concentration type (F,A,T,D,L,U) must still be coded in this field along with NR.

### **5b. Bioconcentration Value (BCF)**

(invoked when BCF/BCFD or BAF selected for Endpoint)

The bioconcentration factor (BCF) is a value, usually reported with units of L/kg, describing the degree to which a chemical can be concentrated in the tissues of an organism in the aquatic environment. At apparent equilibrium during the uptake phase of a bioconcentration test, the BCF is the concentration of a chemical in one or more tissues of the aquatic organism divided by the average exposure concentration in the water. The unitless number is calculated by dividing the concentration of the exposure chemical found in the tissue by the concentration of the chemical found in the exposure water,

$$BCF = \frac{\text{g/kg chemical in organism tissue}}{\text{g/L chemical in H}_2\text{O}}$$

or it is calculated from a ratio of rate constants, if at steady state,

$$BCF = \frac{K1 (\text{uptake})}{K2 (\text{elimination})}$$

A bioconcentration endpoint is coded as either wet (or unknown) or as dry weight (BCF and BCFD, respectively). An accumulation (ACC) effect, measurement of RSDE, and the associated BCF value is coded in the BCF field. If the author does not calculate a BCF, the test is recorded as an ACC effect, measurement of RSDE, NR in the ENDPOINT field, and NR in the BCF field.

If a BCF is reported for the parent compound and for a metabolite, record the parent compound BCF and note "metabolite BCF" in OTHER EFCT.

If the BCF is normalized for lipid, "lipid normalized" is reported in the ENDPOINT REMARK field.

ENDPOINT REMARK: lipid normalized

If an author reports more than one type of BCF, ie. lipid normalized, regular, or radioactive equivalents, for the same data point; code lipid normalized over regular and regular over radioactive equivalents. The secondary analysis endpoint is reported in OTHER EFCT.

OTHER EFCT: radioactive equivalent BCF

For papers that report BCFs and provide Lethal Body Burden information, note “Lethal Body Burden” in OTHER EFCT. However, in a publication reporting only residue data as lethal body burdens code the effect as ACC, the measurement as RSDE, and report “Lethal Body Burden” in MEASUREMENT REMARK.

If an author indirectly measures the uptake of a chemical in an organism by measuring the loss of the chemical from the test media, the data are not coded. However, if the chemical is measured in the excrement (urine or feces) it is coded as RSDE in urine (UR) or feces (FC).

**NOTE:** BCFs of less than 1 (when converted to the standard unit of L/kg) and negative BCF values are suspect. The BCFs less than 1, in the standard unit of L/kg, are coded as reported by the author and negative BCF values are coded as <1. If the value is <1 and not in the standard unit, forward to the EPA database coordinator for review and approval.

If a BCF is reported for an author reported “control” the test set up must be looked at closely. If the test is conducted where a radiolabelled chemical is added to all experimental units and the non-radiolabelled chemical is added to all but one (authors may call this the control), and the authors do not report a clean control (nothing added), the test is then coded with Z control (no control reported). See ECOREF 9212 for an example.

### Statistical Significance (SIG)

The statistical significance field is coded when the author has presented statistical analysis as compared to the controls in the test result.

The valid codes for this field are:

<u>CODE</u>	<u>DEFINITION</u>
SIG	Concentration(s) identified as significant (code lowest concentration in series of significant treatments)
ASIG	All toxicant concentrations significant (including single concentrations exposures)
NOSIG	Concentrations identified as not significant (code highest concentration in series of non-significant treatments)
ANOSIG	All toxicant concentrations not significant (including single concentrations exposures)
MULT	Combination of sig and nosig results; pattern of significance is non-monotonical, or results combined due coding practices.
NA	Not applicable (use for LC50, EC50, BCF, MATC, NR-LETH, AND NR-ZERO)
NR	Not reported

If statistics are presented in the publication, unless the authors state otherwise, assume that the exposure treatments were compared to the controls.

When statistical comparisons are presented for multiple controls (e.g., statistics in relation to a standard control and statistics in relation to a solvent control), code M in the

CONTROL TYPE field and note the appropriate control type used for the coded statistics in MEASUREMENT REMARK. (e.g., "stats based on solvent control") The data related to the solvent control is coded preferentially over the standard control data and should be noted as such in the MEASUREMENT REMARK field.

Data are separated into individual records if statistics are based on concentrations and a clear dose response is shown. For example:

- 1) If only one concentration is presented and the response differs over time, code four records

Example 1:

Conc.	24h	48h	72h	96h
1 ug/l	nosig	nosig	sig	sig

Code four Endpoints : 2 NOECs at 24 and 48 hours and 2 LOECs at 72 and 96  
Hours at CONC: 1 ug/l SIGNIF for NOECs : ANOSIG, SIGNIF for LOECs: ASIG

- 2) If multiple concentrations and durations are presented. Code the statistics at each duration separately.

Example 2:

Conc.	24h	48h	72h	96h
1 ug/l	nosig	nosig	nosig	sig
2 ug/l	nosig	nosig	sig	sig
3 ug/l	nosig	sig	sig	sig

Code: NOEC at 24 h 3 ug/L; NOEC/LOEC at 48h 2 ug/L 3 ug/L; NOEC/LOEC at 72 h 1 ug/L 2 ug/L; LOEC at 96 h 1 ug/L

Use the Companion Endpoint tab to mark for each NOSIG SIG companion endpoint group

- 3) If multiple concentrations and durations are presented and results are non-monotonic within each duration code one record combining all concentrations all durations.

Example 3:

Conc.	24h	48h	72h
1 ug/l	nosig	sig	nosig
2 ug/l	sig	sig	sig
3 ug/l	nosig	nosig	nosig

Code Conc: 1-3 ug/L, TIME: 24 h, SIGNIF: Mult ENDPOINT: NR  
Conc: 1-3 ug/L, TIME: 48 h, SIGNIF: Mult ENDPOINT: NR  
Conc: 1-3 ug/L, TIME: 72 h, SIGNIF: Mult ENDPOINT: NR

The SIG field is coded as "NA" for records having an endpoint of MATC, LCxx, ECxx, LTxx, BCF, ETxx, ICxx, LDxx, LETC, BCFD, NR-LETH, and NR-ZERO. For effects without endpoints, code significance as author reports, or NR.

The significance for LOEC and NOEC must be coded as SIG and NOSIG respectively unless a publication reports a LOEC and NOEC for a non-monotonical response pattern (i.e., lower concentration significant and at least one higher concentration not

significant); In this case code the LOEC/NOEC reported by the authors, but note TREND as 'CHG' and code SIG as 'MULT' to flag non-standard results.

The reviewer interprets hypotheses tests to determine a dose response endpoint. A significant clear dose result is coded as a LOEC; no significant dose result is coded as a NOEC. Only the highest NOEC (NOSIG) and the lowest LOEC (SIG) concentration is reported; unless all concentrations are SIG or all concentrations are NOSIG. In this instance, code the NOEC/LOEC as identified and note that the significance is ASIG (all significant) or ANOSIG (all not significant) respectively. If the significance level is reported, it is coded in the LEVEL field described below.

In cases where the author reports only a LOEC (SIG) or NOEC (NOSIG), code the companion data point. For example, if a stat sig "growth" is reported in the text and in the table sig is noted the reviewer should pick the nosig level and report this also.

If the author states that there is a statistically significant increase or decrease in an observed effect, whether or not they report the statistical method used, but does not report a significance level, code SIG or NOSIG and NR in LEVEL field.

If a table has a footnote defining \* values as significant as  $p < xx$ , it is acceptable for the reviewer to assume that data points without an asterisk are not significant.

If the author states there is a significant increase or decrease in an observed effect but does not say it is "statistically significant," code NR in SIG field.

- When the highest concentration and all lower concentrations tested show no significant response, code NOEC ANOSIG and the highest concentration in CONC field.

Example:      10 ug/L    NOSIG    P<0.05  
                  20 ug/L    NOSIG    P<0.05  
                  30 ug/L    NOSIG    P<0.05

Code:    Endpoint:    NOEC    CONC: 30 ug/L      SIG: ANOSIG      LEVEL: P<0.05

- When the lowest concentration and all higher concentrations tested show a significant response, code LOEC ASIG and the lowest concentration in CONC field.

Example:      10 ug/L      SIG      P<0.05  
                  20 ug/L      SIG      P<0.05  
                  30 ug/L      SIG      P<0.05

Code:      Endpoint:    LOEC    CONC: 10ug/L      SIG: ASIG    LEVEL: P<0.05

- If only one concentration is tested and statistics are performed, code ASIG or ANOSIG in stats and "only conc tested" as a CONC remark.

Example:      10 ug/L SIG      P<0.05

Code:              Endpoint:    LOEC    CONC: 10ug/L      ASIG      P<0.05    Conc REMARK: "only conc tested"

If an author reports p values but the level at which the data are considered significant is not reported by the author, use the conservative significance level of  $P \leq 0.05$  as a



benchmark. The ENDPOINT ASSIGNED is coded as R and ENDPOINT REMARK is coded with "SIG ASSIGNED"

Example: The paper reports 9 unique mRNA measurements at one concentration and gives p values but does not report the level that is considered significant. Using a benchmark level of significance of  $p \leq 0.05$  the following endpoints are coded:

mRNA Name	P VALUE	ENDPOINT ASSIGNED	ENDPOINT	ENDPOINT REMARK
BHMT	0.57	R	NOEC	SIG ASSIGNED
ETIF	0.04	R	LOEC	SIG ASSIGNED
UCE	0.01	R	LOEC	SIG ASSIGNED
Tubulin	0.21	R	NOEC	SIG ASSIGNED
Trypsin	0.07	R	NOEC	SIG ASSIGNED
UBI	0.14	R	NOEC	SIG ASSIGNED
SAMS	0.32	R	NOEC	SIG ASSIGNED
Hcy a	0.72	R	NOEC	SIG ASSIGNED
Hcy b	0.31	R	NOEC	SIG ASSIGNED

If a paper presents statistically analyzed data using least square difference (LSD) analysis, determine the statistical significance by the following calculation:

Dose	Response
Control 1	2.4
Dose 2	4.6
Dose 3	4.8
Dose 4	6.7
LSD(0.05)	1.1

Subtract or add the LSD value (1.1) to the control value to get the lowest value that is significantly different. In this example, anything above 3.5 or below 1.3 is a significant value at the  $p=0.05$  level. Therefore, all doses are significantly different from the control because they are all greater than 3.5 (Pers. Comm., R. Regal, UMD Statistics Dept., 1999).

Combining of Statistics

If a measurement has no clear dose response as interpreted by the reviewer when statistics are reported, it is coded as multiple significance (MULT).

Example 1: Five concentrations are tested and the two highest and two lowest show significance but the middle concentration does not, code MULT.

Growth table with length affected by copper.

Conc	Control	1 ug/L	2 ug/L	3 ug/L	4 ug/L	5ug/L
Length	20 cm	35cm	36cm	21cm	15cm	14cm
		sig	sig	nosig	sig	sig
						P<0.05

Code: EFCT: GRO MEASURE: LGTH SIGNIF: MULT LEVEL: p<0.05  
 CONC: 1-5 ug/L ENDPOINT: NR

Example 2: A publication reports a LOEC and NOEC for a non-monotonical response pattern (i.e., lower concentration significant and at least one higher concentration not significant); code the LOEC/NOEC reported by the authors, note TREND as 'CHG', and code SIG as 'MULT' to flag non-standard results.

Mortality	Control	10 ug/L	20 ug/L	30 ug/L	40 ug/L	50 ug/L
@ 24 hours		nosig	nosig	sig	nosig	sig

Author endpoints NOEC LOEC

Code: ENDPT: NOEC EFCT: MOR MEASURE: MORT  
 CONC: 20 ug/L SIGNIF: MULT TREND: CHG

ENDPT: LOEC EFCT: MOR MEASURE: MORT  
 CONC: 30 ug/L SIGNIF: MULT TREND: CHG

**Note:** The MULT code is not used to represent a combination of data which has been statistically analyzed with data that have not been analyzed. Code the statistical data over the non-statistically analyzed data. For example, in an instance where the exposure duration is 5 days, and the statistical results presented are for 4 days; code the statistical results, the observed duration as 4 days and enter the exposure duration as 5 days.

Significance Level (LEVEL)

The level of significance (e.g. test statistic) is coded when the author has reported statistical analysis in the test result. The terminology for significance level may be presented as: p =; p- or alpha value;  $\chi^2$ ; for t-test; % level. The terminology are equivalent and are generally in the range of p = 0.10 to p = 0.001.

The LEVEL field is coded as "NA" for records having an endpoint of MATC, LCxx, ECxx, LTxx, BCF, ETxx, ICxx, LDxx, LETC, BCFD, NR-LETH, and NR-ZERO. However, when the confidence level is other than 95%, the level is coded as reported.

If an author reports two levels of statistical significance (e.g. 0.05 and 0.01, 5% and 1%) for the same data point and the significance is different at the levels, the level at which the data is first statistically significant is coded. For example:

Premature Molting for 15 days is analyzed at 5% and 1%

Concentration (ug/L)	% premature molting	5% , 1% *	Code
control	0	d , C	LOEC at 5% significance at 1 ug/L
1	15.38	c, C	
30	66.67	b, B	
60	92.59	a, AB	
100	90.00	a, AB	
200	96.67	a, A	

\* Same characters in a column indicate no significant difference

If an author reports two levels of statistical significance (e.g. 0.05 and 0.01, 5% and 1%) for the same data point and the significance is the same at both levels, the most sensitive level (e.g. 0.01) at which the data is first statistically significant is coded. For example:

Mortality for 9 days is analyzed at p=0.05 and p=0.01

Concentration (ug/L)	% Mortality	P=0.05 , p=0.01 *	Code
control	6.3	b , B	NOEC at p=0.01 at 25 ug/L, LOEC at p=0.01 at 50 ug/L
25	6.4	b, B	
50	24.6	a, A	
100	20.2	a, A	
150	32.2	a, A	

\* Same characters in a column indicate no significant difference

### Combining of Level

When a range of concentrations is coded, and there are multiple levels of significance reported, range the values.

Non-monotonical response was reported for data with concentrations of 15, 30 and 60 ug/L. At 15 ug/L the p-value was p<0.05 (sig), at 30 ug/L the p-value was p<0.10 (nosig) and at 60 ug/L the p-value was p<0.001 (sig).

CONC: 15 to 60    SIG: Mult    LEVEL: p<0.10 to 0.001    ENDPOINT: NR

### Chemical Analysis Method (M/U)

This parameter identifies whether quantitative analyses of the toxicant concentration in the test media were conducted and whether measured concentrations were used to report the results.

This field represents/defines the concentration which was used in reporting the endpoint or effect; publications may report **M**easured and **U**nmeasured concentrations for one test scenario, use the code which represents whether the specific effect/endpoint concentration was measured or unmeasured. If both measured and unmeasured concentrations for the specific effect/endpoint are reported, record only the measured concentrations.

For instances where some treatment (dose) levels are measured and some are unmeasured/nominal, and all unmeasured/nominal concentrations are reported, report the unmeasured/nominal concentrations for each treatment level so that the range of concentrations is consistent and monotonical. Denote all of the concentration analysis methods as 'X', signifying that a mixture of measured and unmeasured values was reported.

When chemical measurements are conducted on stock solutions, but nominal concentrations are reported for effects or endpoints, code as **U**nmeasured. When chemical measurements are conducted periodically throughout the exposure but the reported measurements are not correlated with the effects, code as **U**nmeasured. When chemical measurements are conducted periodically throughout the exposure and the effects are coordinated with the measurements, code as **M**easured. For non-English publications, code as **N**ot **R**eported unless explicitly stated to be measured or unmeasured concentrations.

Even if measured values are reported by the author to have deteriorated by the end of the exposure, the **M**easured code should still be used. It is acceptable to assume that if the author used measured concentrations in residue analysis, that these measured values were carried over to calculate BCFs. (See ECOTOX Appendix P for codes)

### Assigned Endpoint (ASSIGNED ENDPOINT)

If the reviewer codes an endpoint which is not stated by the author in the paper, or interprets an author's endpoint definition to be equivalent to the ECOTOX endpoint definition, an "**R**" for reviewer-assigned endpoint is entered into the field. In all cases except for NR-ZERO and NR-LETH, "by definition", the endpoint the author reports or the description of the endpoint is coded in the ENDPOINT REMARK data field. However, if the reviewer codes an endpoint as the author states it in the paper, a "**P**" is coded for paper-assigned endpoint.

**Note:** An "**R**" is always coded for the endpoints NR-ZERO and NR-LETH, however no further information is coded in the ENDPOINT REMARK field.

Reviewers will follow these guidelines in assigning endpoints:

1. BY DEFINITION: If the author does not actually state that the value is an LD50 but states that "concentration x is the dose estimated to be lethal to 50% of the test organisms", the reviewer should code this as an LD50 endpoint because the author defines the LD50. Such a designation is accompanied by noting "**R**" in the PR data field.

2. When the author provides text which identifies a value as the “highest tested concentration having no statistically significant adverse effect”, the reviewer should code this as a NOEL/NOEC; the “lowest tested concentration having a statistically significant adverse effect” is coded as a LOEL/LOEC. In both cases, the PR field will be coded as 'R' to reflect reviewer assignment of an endpoint. Because LOEL/NOEL values are assigned under very specific experimental and statistical conditions, ECOTOX reviewers will be assigning complementary NOELs or LOELs only when the author assigns either a LOEL or NOEL.

3. When the author provides statistical information, which designates concentrations as significantly different from the control, the reviewer will code this information as SIG or NOSIG. The reviewer will also report the level of significance in the LEVEL data p-value in the p-value field.

4. An author reports an Endpoint similar to one defined in the ECOTOX SOP.

Example 1. Author uses TLM for 50% mortality endpoint. Reviewer can code the endpoint as LC50 and puts “**R**” for reviewer assigned endpoint (code TLM in the ENDPOINT REMARK field).

5. Coding a companion endpoint

Example 2. Author reports NOEC in paper but does not include companion endpoint - LOEC. If reviewer can determine LOEC from data, an “**R**” is coded for the LOEC (code “by definition” in the ENDPOINT REMARK field) and a “**P**” is coded for the NOEC.

6. If the author presents biologically significant data which does not include statistical analysis because all of the animals died, the reviewer should not interpret this data as a non-monotonical response. Code the appropriate biologically significant endpoints as NOEC/LOEC values. For example:

Example 3

WHOLE BODY GROWTH

CONTROL	139 G
10 UG/ML	138 G
20 UG/ML	136 G
30 UG/ML	100 G*
40 UG/ML	95 G*
50 UG/ML	ALL DEAD

\* = SIGNIFICANT AT 0.05

CODE: EFFECT: GRO (GROWTH)  
MEASURE: WGHT (WEIGHT)

ENDPOINTS: NOEC @ 20 UG/ML  
LOEC @ 30 UG/ML

Example 4

% DEFORMITY

CONTROL	0%
10 UG/ML	0%
20 UG/ML	0%
30 UG/ML	0%
40 UG/ML	100%
50 UG/ML	ALL DEAD

CODE: EFFECT: DVP (DEVELOPMENT)  
MEASURE: DFRM (DEFORMATION)

ENDPOINTS: NOEC @ 30 UG/ML  
LOEC @ 40 UG/ML

Weight at time of result (WTAT)

The body weight of the organism at the time of the observed effect response is reported. If multiple body weights are given throughout the test, report the weight at the time the response is reported. If a range of weight is given for the organisms, report the range of weights. Report the treatment group weight, if weights are given for both a treatment group and a control group.

Intake Rate (INTAKE)

Report the intake rate of the exposure media (not the contaminant) at the time of the result (or closest rate to that time) for diet studies (through food or drinking water) as reported by the author.

Test Result Examples

1. If the author has defined an ENDPOINT and/or has reported a 0% and/or 100% mortality response, report the endpoint/mortality as outlined in the Endpoint Hierarchy. Select the appropriate effect as described below.

ENDPOINT REPORTED (NR-ZERO):

ENDPOINT: NR-ZERO	MEASURE: MORT	
TREND: NEF	EFCT%: 0	
EFFECT: MOR	SIG: NA	LEVEL: NA

If applicable, statistical results should appear in the SIG field, the level of significance should be reported in the LEVEL field, the percent effect should be presented in the EFCT% field, and the trend should be reported in the TREND field.

ENDPOINT REPORTED (LOEC):

ENDPOINT: LOEC	MEASURE: LGTH	
TREND: DEC	EFCT%: 20	
EFFECT: GRO	SIG: SIG	LEVEL: $\alpha < 0.05$

**Note:** For NOEC endpoints, NOSIG is coded in the SIG field. For LOEC endpoints, SIG is coded in the SIG field.

2. If the author-reported effect is a clear dose response result using statistical analysis, and the author does not identify an endpoint, select the appropriate effect from ECOTOX Appendix S.

Clear dose response data where a statistically significant effect was observed, are represented by two data records. One data record represents the lowest concentration at which a statistically significant effect occurred. An Endpoint, LOEC is encoded and "SIG" is coded in the SIG field, the observed trend is coded in the TREND field, the percent effect is coded in the EFCT% field, and the level of significance is reported in the LEVEL field. The observed measurement is coded in the MEASURE field. Remarks on the effect are made in the EFFECT\_REMARK field.

CLEAR DOSE RESPONSE:

ENDPOINT: LOEC	SIGNIF: SIG	TREND: DEC
EFFECT: GRO	EFCT%: 20	LEVEL: $\alpha < 0.05$
MEASURE: LGTH		

The second data record represents the highest concentration at which no effect occurred. An Endpoint, NOEC is encoded and NOSIG is coded in the SIG field. If a percent effect is reported it is presented in the EFCT% field.

If the concentration identified as SIG is the lowest concentration reported or the concentration identified as NOSIG is the highest concentration reported, code the NOEC/LOEC at the highest/lowest concentration and the appropriate code (ASIG and ANOSIG) in the SIG field.

Standard methods recommend that when determining a NOEL/LOEL, at least three exposure concentrations be used (Menzer 1994 at 1406);. If the author uses only one exposure concentration AND assigns a NOEL/LOEL or SIG/NOSIG result, a Remark noting "only conc tested" will be coded.

3. If the author reported effect shows unclear dose response results, using statistical analysis, select the appropriate effect from ECOTOX Appendix S.

When data have been statistically analyzed, and the results presented have significant effects in an unclear dose response pattern (e.g., significant effects at the high and low concentrations, and not significant at the middle concentration), "MULT" is coded in the SIG field to signify multiple significance. The level is coded with a full range of p-values (e.g.  $p < 0.05$ -0.001).

UNCLEAR DOSE RESPONSE:

ENDPOINT: NR	MEASURE: ACHE	
TREND: CHG		
EFFECT: ENZ	SIG: MULT	LEVEL: $P < 0.05$ -0.001

4. If the author reports a descriptive or qualitative effect without statistical analysis, select the most appropriate effect from ECOTOX Appendix S. One record is coded with a full range of exposure concentration and time. Code all measurements for the specific effect. The appropriate trend is coded in the TREND field. If these data report multiple trends, code CHG in the TREND field and report the individual trends in MEASUREMENT REMARK field. The percent effect over the concentration tested is reported in the EFCT% field. NR is coded in SIG and LEVEL fields.

NO STATISTICAL ANALYSIS:

ENDPOINT: NR	MEASURE: LESI, ATRS, DEGN, EDMA, EXCS	MEASUREMENT REMARK: INC LESI, ATRS, DEGN, EDMA, DEC EXCS
TREND: INC	EFCT%: NR	
EFFECT: HIS	SIG: NR	LEVEL: NR

### Percent Lipid (LD)

If percent lipid information is provided in the publication, record as a % value

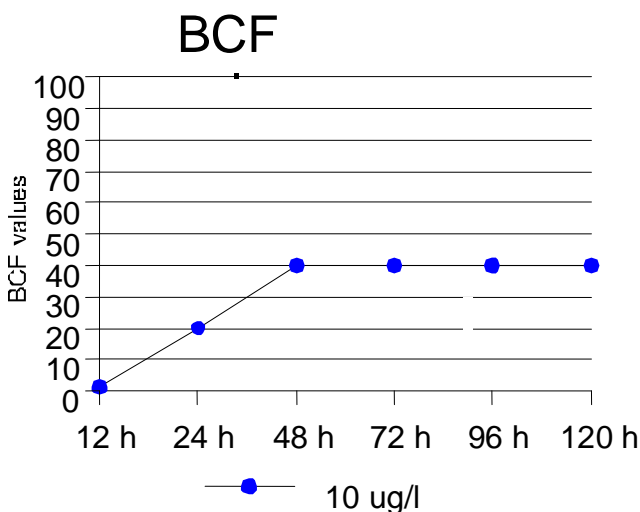


Dry or Wet Weight and Percent (DW)

Record whether the residue/bioconcentration/bioaccumulation, growth, population or biochemical data are reported on a dry or wet weight basis , by selecting D, W or NR from the dropdown. If percent moisture is reported, record the percentage value . If the author does not report this information code as 'NR'.

Steady State (STST)

Record whether the residue/bioconcentration/bioaccumulation data at the time of the result is at steady state or equilibrium by selecting Yes, No or NR from the dropdown.. If the author does not report in the text that the data are at steady state or equilibrium but represents the data graphically, this information can be determined by the graphed data. If the residue or BCF data are presented in a graph and the data points flatten out and continue to flatten during the exposure phase, the point at which it flattens out is the steady state or equilibrium point. For Example:



The steady state or equilibrium is reached at 48 hours. STST is coded as YES for the BCF values at 48, 72, 96 and 120 hours.

If the author does not report this information code As 'NR'

The steady state or equilibrium is reached at 48 hours. STST is coded as YES for the BCF values at 48, 72, 96 and 120 hours.

If the author does not report this

information code NR.

**Result Remarks**

The Result Remarks data field contains historical comments from both the aquatic and terrestrial databases. No entries are made in the Result Remarks data field. If records are completed for data maintenance, the comments in the Result Remarks data field will be moved to the corresponding remark data field.

## 6. Media Characteristic Parameters tab

**Figure 7. Media Characteristic tab**

The following media/water chemistry parameters are included in ECOTOX, and are coded in appropriate fields.

These measured values pertain either to the test media/water chemistry or the dilution or culture media/water chemistry values. In the absence of test media/water chemistry parameters, it is acceptable to report the culture, holding tank, acclimation, control or dilution media/water, or pretest conditions denoted by an asterisk (\*) which is entered directly after the value (e.g. 8.6\* mg/L). Media/water chemistry parameters measured prior to or after the exposure period are coded only if test media/water chemistries are not reported in the publication. If the author reports the test conditions as “similar” to other methods in the paper, code test conditions as “NR”.

When media/water chemistries differ between samples (e.g., test chamber or water body), and results are obtained from only some of the samples, media/water chemistries should be reported for only those samples actually tested. The following are two examples on coding media/water chemistries:

Conc	Temperature (Celsius)	pH
1 ug/l	20.4 (20.1 - 20.7)	7.9 (7.5 - 8.3)
2 ug/l	20.5 (20.2 - 20.8)	8.0 (7.6 - 8.4)
3 ug/l	20.4 (20.0 - 20.8)	7.8 (7.7 - 7.9)

Example 1: Statistics are presented for Protein content and it is found to be significant at 2 ug/l. The data coded for Temperature and pH are for the specific concentration.

Code: Endpoint: LOEC Effect: BCM Measurement: PRCO Concentration: 2 ug/l Significance: SIG pH: 8.0 (7.6-8.4) Temperature: 20.5 (20.2-20.8)

Example 2: Statistics are presented for Glucose content and all concentrations are found to be not significant. The data coded for Temperature and pH are for the range of concentrations.

Code: Endpoint: NOEC Effect: BCM Measurement: GLUC Concentration:3 ug/l Significance:  
ANOSIG pH: 7.5-8.4 Temperature: 20.0 - 20.8

When the author refers to the media/water chemistry values as approximate a "~" is coded in front of the value.

Graphed data are coded as a range or as "less than" or "greater than" values and the term "graphed" is noted in the corresponding remark data field.

Media/water chemistry values should be coded as reported by the author.

For studies where a chemical concentration is monitored as a water or soil chemistry parameter (e.g. sodium chloride reported as conductivity, calcium chloride reported as hardness), code the water or soil chemistry parameter in both the dose and the water chemistry parameter data fields.

#### Aquatic Studies Media Parameters

(invoked by selecting 'Water' from the Species Habitat data field)

Field Name	Standard Unit	Abbreviation	Comment
Temperature	C	<u>TEMP</u>	When temperatures are reported for incubation chambers or water baths, these temperatures are acceptable for reporting as test temperatures. Do not code temperatures noted as "room temperature".
Hardness	mg / L CaCO <sub>3</sub>	<u>HARD</u>	If the author only reports the terms "hard" or "soft", these terms are recorded. If the author reports a hardness value but does not identify a unit and/or refers to the value as "total", standard units are assumed and the value coded.
Alkalinity	mg / L CaCO <sub>3</sub>	<u>ALK</u>	If the author reports an alkalinity value but does not identify a unit and/or refers to the value as "total", standard units are assumed and the value coded.
Dissolved Oxygen	mg / L	<u>DO</u>	A "SAT" code is used for 100% saturation
pH	--	<u>pH</u>	pH range is between 1 and 14
Salinity	0/00	<u>SALIN</u>	Some authors report the unit as ppt. This is parts per thousand and should be coded as 0/00 not as parts per trillion ppt.
Conductivity	umhos / cm	<u>COND</u>	
Organic Carbon	mg / L Carbon	<u>ORG C</u>	Organic carbon must designate (T=Total, P=Particulate, D=Dissolved); if more than one type of organic carbon is reported in the publication, record T in the field and the other values (P or D) as a Remark; if the value is reported as "organic carbon" without identifying type, assume the value is expressed as Total

Field Name	Standard Unit	Abbreviation	Comment
			and report T. Sediment organic carbon values are not reported.
Humic Acid	%	<u>HA</u>	
<u>Sodium</u>	mg/l	<u>Na</u>	
<u>Chlorine</u>	mg/l	<u>Cl</u>	
Dissolved inorganic carbon	mg/l	<u>DIC</u>	
Calcium	mg/l	<u>Ca</u>	
Potassium	mg/l	<u>K</u>	
Magnesium	mg/l	<u>Mg</u>	
Sulfate	mg/l	<u>SO4</u>	
Sulfur	mg/l	<u>S</u>	

### Terrestrial Studies Media Parameters

(invoked by selecting 'Soil' from the Species Habitat data field)

Field Name	Standard Unit	Abbreviation	Comment
Temperature	C	<u>TEMP</u>	When temperatures are reported for incubation chambers or water baths, these temperatures are acceptable for reporting as test temperatures. Do not code temperatures noted as "room temperature".
Soil Texture Sand	%	<u>SA</u>	Report the sand texture of the soil as stated by the authors
Soil Texture Silt	%	<u>SI</u>	Report the silt texture of the soil as stated by the authors
Soil Texture Clay	%	<u>CL</u>	Report the clay texture of the soil as stated by the authors Clay may be reported as bentonite, kaolinite or montmorillonite.
CEC	uS/cm	<u>CEC</u>	Report cation exchange capacity and units (Refer to Appendix FF for cation exchange capacity units) of the test media.
pH	--	<u>pH</u>	pH range is between 1 and 14 If the author specifies a measurement method for the pH value (e.g., that the pH value is measured by pHKCl or pHCaCl2), code the pH value and identify the measurement method in the REMARKS field. If the authors report that a standard (see Attachment G for list of standard soils) or commercially available artificial soil is used, but do not present pH, use the pH reported in the standard test method referenced by the author. If the pH of a specific soil type is not given in the

Field Name	Standard Unit	Abbreviation	Comment
			publication, a search of the USDA/NRCS National Cooperative Soil Survey (USA) online site, at the following web address: <a href="http://ncsslslabdatamart.sc.egov.usda.gov/querypage.aspx">http://ncsslslabdatamart.sc.egov.usda.gov/querypage.aspx</a> can be conducted for the Characterization Data of the specific soil series. If the pH is found, range the pH values for all soil depths in the pH data field and remark in the comments section "from USDA web source". Attach a printout of the pH information from this site to the publication.
Organic Matter	mg / L	<u>OM</u>	If the authors report that a standard (see Attachment G for list of standard soils) or commercially available artificial soil is used, but do not present organic matter content, use the organic matter content reported in the standard test method referenced by the author.  If the organic matter of a specific soil type is not given in the publication, a search of the USDA/NRCS National Cooperative Soil Survey (USA) online site, at the following web address <a href="http://ncsslslabdatamart.sc.egov.usda.gov/querypage.aspx">http://ncsslslabdatamart.sc.egov.usda.gov/querypage.aspx</a> can be conducted for the Characterization Data of the specific soil series. If the organic matter is found, range the organic matter values for all soil depths in the OM data field and remark in the comments section "from USDA web source". Attach a printout of the organic matter information from this site to the publication.
Moisture	%	<u>MOIST</u>	

For Terrestrial Non-Soil Studies, only the Temperature data fields will be invoked. If pH of a culturing media, e.g. oocyte rearing solution, is reported, encode the pH in the Experimental Design data field.

## 7. Dose Response tab

Dose Response data is not being coded at this time.

Response data for each dose tested is reported by encoding each individual data point, e.g. percent mortality, number of offspring, amount of protein, etc.

Select the Result record that is associated with the Dose Response data to be coded. Select Add Dose Response button to enable the dose response screen entry fields.

ID	Sample Unit	Measurement	Response Site	Duration	0 mg/kg (NC) C	100 mg/kg (NC)	500 mg/kg (NC)	1000 mg/kg (NC)	5000 mg/kg (NC)	10000 mg/kg (NC)	Collapse
52684	13510491 - Beryllium sulfate	Non-Sol	4991 - Elsentia fetida	NR	AD	NC	UA	LAB	5	12/10/2002	07/20/2012
52685	7446324 - Sulfuric acid antimony(3+) salt (3:3)	Non-Sol	4991 - Elsentia fetida	NR	AD	NC	DA	LAB	5	12/10/2002	07/20/2012
52686	7446324 - Sulfuric acid antimony(3+) salt (3:3)	Non-Sol	4991 - Elsentia fetida	NR	AD	NC	DA	LAB	5	12/10/2002	07/20/2012
52687	7785877 - Sulfuric acid, Manganese(2+) salt (1:1)	Non-Sol	4991 - Elsentia fetida	NR	AD	NC	DA	LAB	1	12/10/2002	07/20/2012
52688	7785877 - Sulfuric acid, Manganese(2+) salt (1:1)	Non-Sol	4991 - Elsentia fetida	NR	AD	NC	DA	LAB	5	12/10/2002	07/20/2012
52689	7785877 - Sulfuric acid, Manganese(2+) salt (1:1)	Non-Sol	4991 - Elsentia fetida	NR	AD	NC	DA	LAB	5	12/10/2002	07/20/2012
52690	7785877 - Sulfuric acid, Manganese(2+) salt (1:1)	Non-Sol	4991 - Elsentia fetida	NR	AD	NC	DA	LAB	1	12/10/2002	07/20/2012
52691	13510491 - Beryllium sulfate	Non-Sol	4991 - Elsentia fetida	NR	AD	NC	DA	LAB	1	12/10/2002	07/20/2012
52692	13510491 - Beryllium sulfate	Non-Sol	4991 - Elsentia fetida	NR	AD	NC	DA	LAB	5	12/10/2002	07/20/2012

Dose Response Information	
Sample Unit	Dose Response Unit
AD	0%
Duration	14 d
Mean	Min
Max	Unit
Comments	Replicate 1

ID	Dose (Conc Type)	Dose Response	Stat. Method	Stat. Method Value	Sample Size	Significance	Significance Level	Details												
73465	0 mg/kg (NC) C	5	NR	NR	5	NR	NR	Details												
<table border="1"> <thead> <tr> <th>Dry Wet</th> <th>Dry Wet Percent</th> <th>Percent Lipid</th> <th>Comment</th> </tr> </thead> <tbody> <tr> <td>NR</td> <td>NR</td> <td>NR</td> <td></td> </tr> <tr> <td>Mean</td> <td>Min</td> <td>Max</td> <td>Comments</td> </tr> </tbody> </table>									Dry Wet	Dry Wet Percent	Percent Lipid	Comment	NR	NR	NR		Mean	Min	Max	Comments
Dry Wet	Dry Wet Percent	Percent Lipid	Comment																	
NR	NR	NR																		
Mean	Min	Max	Comments																	
73466	100 mg/kg (NC)	5	NR	NR	5	NR	NR	Details												
<table border="1"> <thead> <tr> <th>Dry Wet</th> <th>Dry Wet Percent</th> <th>Percent Lipid</th> <th>Comment</th> </tr> </thead> <tbody> <tr> <td>NR</td> <td>NR</td> <td>NR</td> <td></td> </tr> <tr> <td>Mean</td> <td>Min</td> <td>Max</td> <td>Comments</td> </tr> </tbody> </table>									Dry Wet	Dry Wet Percent	Percent Lipid	Comment	NR	NR	NR		Mean	Min	Max	Comments
Dry Wet	Dry Wet Percent	Percent Lipid	Comment																	
NR	NR	NR																		
Mean	Min	Max	Comments																	
73467	500 mg/kg (NC)	5	NR	NR	5	NR	NR	Details												
<table border="1"> <thead> <tr> <th>Dry Wet</th> <th>Dry Wet Percent</th> <th>Percent Lipid</th> <th>Comment</th> </tr> </thead> <tbody> <tr> <td>NR</td> <td>NR</td> <td>NR</td> <td></td> </tr> <tr> <td>Mean</td> <td>Min</td> <td>Max</td> <td>Comments</td> </tr> </tbody> </table>									Dry Wet	Dry Wet Percent	Percent Lipid	Comment	NR	NR	NR		Mean	Min	Max	Comments
Dry Wet	Dry Wet Percent	Percent Lipid	Comment																	
NR	NR	NR																		
Mean	Min	Max	Comments																	
73468	1000 mg/kg (NC)	5	NR	NR	5	NR	NR	Details												
<table border="1"> <thead> <tr> <th>Dry Wet</th> <th>Dry Wet Percent</th> <th>Percent Lipid</th> <th>Comment</th> </tr> </thead> <tbody> <tr> <td>NR</td> <td>NR</td> <td>NR</td> <td></td> </tr> <tr> <td>Mean</td> <td>Min</td> <td>Max</td> <td>Comments</td> </tr> </tbody> </table>									Dry Wet	Dry Wet Percent	Percent Lipid	Comment	NR	NR	NR		Mean	Min	Max	Comments
Dry Wet	Dry Wet Percent	Percent Lipid	Comment																	
NR	NR	NR																		
Mean	Min	Max	Comments																	

**Figure 8. Dose Response tab**

### Sample Unit

Code as described above.

### Measurement

Code as described above.

### Response Site

Code as described above.

### Dose Response Unit

Enter the Unit of the Dose response observed.

### Observation Duration

Code as described above.

### Dose Response Comment

Enter the comment for all dose records.

Dose Response Value, Stat Method and Stat Method Value Enter the greater than (>), less than (<), minus (-) or approximation (~) symbols, if reported, as used by the author(s) to describe the response value preceding the MEAN or RANGE data field entries.

Report the mean or single observed response value, as reported in the publication, in the MEAN data field. When individual response values are reported along with a sum of all values, report each individual response value as well as the sum/total value on separate result lines.

Report the range or confidence (or fiducial) intervals (or limits) which are defined as CL (confidence limit), CI (confidence interval), FI (fiducial interval), or FL (fiducial limit) of the response value in the RANGE data field. The type of data stored in the RANGE data field will be identified in the SM data field (e.g., Data reported as a range (with a mean) or confidence interval (with an endpoint) will be specifically identified in the SM field. It is also assumed that the confidence interval is calculated at 95% and is noted in the LEVEL data field.

When the measurement unit includes a standard deviation (SD) or standard error (SE), specifically identify these types of ranges in the SM data field. Report the numeric value of the standard deviation or standard error in the VALUE data field.

Report the measurement unit which corresponds to the MEAN and/or RANGE entry in the UNIT data field (see Appendix N for standard units).

#### Sample Size

Code as described above.

#### Significance

Code as described above.

#### Significance Level

Code as described above.

#### Dry/Wet

Code as described above.

#### Dry/Wet Percent

Code as described above.

#### Percent Lipid

Code as described above.

#### Individual Dose Response Comment

Enter the comment related to the specific dose being coded.

### **8. Companion Endpoint(s) tab**

(only invoked if NOxx, LOxx or MATC coded)



Reference

Skim

ECOREF#: 69402

Lethal and Subchronic Effects of 2,4,6-Trinitrotoluene (TNT) on *Enchytraeus albidus* In Spiked Artificial Soil by Dodard, S.G., A.Y. Renoux, J. Pawlowski, and G.J. Sunahara, 2003

Warnings: 1

ID	Chemical	Habitat	Species	Age	Lifestage	Study Type	Exposure Type	Test Location	#Results	Create Date	Modified Date	Collapse	
2052205	118967 - 2,4,6-Trinitrotoluene	Soil	1499 - <i>Enchytraeus albidus</i>	NR	MA	NR	EN	LAB	8	04/22/2013	04/22/2013		
ID	Sample Number	Trend	Endpoint	Effect	Measurement (s)	Response Site(s)	Significance	Observed Duration	Concentration	Page Number	Reviewer Number	Modified Date	Collapse
2087262		INC	LC20	MOR	MORT	NR	NA	21 d	F 234 (147 - 301) mg/kg d soil	135	5	04/22/2013	
2087263		INC	LC50	MOR	MORT	NR	NA	21 d	F 362 (275 - 438) mg/kg d soil	135	6	04/22/2013	
2087264		INC	LOEC (NOEC)	MOR	MORT	NR	SIG	21 d	F 389 mg/kg d soil	135	7	04/22/2013	
2087265		INC	NOEC (LOEC)	MOR	MORT	NR	NOSIG	21 d	F 195 mg/kg d soil	135	8	04/22/2013	
2087266		DEC	EC20	REP	FCND	NR	NA	42 d	F 88 (38 - 115) mg/kg d soil	135	9	04/22/2013	
2087267		DEC	EC50	REP	FCND	NR	NA	42 d	F 133 (95 - 173) mg/kg d soil	135	10	04/22/2013	
2087268		DEC	LOEC (NOEC)	REP	FCND	NR	SIG	42 d	F 194 mg/kg d soil	135	11	04/22/2013	
2087269		DEC	NOEC (LOEC)	REP	FCND	NR	NOSIG	42 d	F 97 mg/kg d soil	135	12	04/22/2013	

Result Information			Media Characteristics (Soil)			Dose Response			Companion Endpoints		
ID	Sample Number	Trend	Endpoint	Effect	Measurement(s)	Response Site(s)	Significance	Observed Duration			
<input checked="" type="checkbox"/> 2087265	NR	INC	NOEC	MOR	MORT	NR	NOSIG	21 d			
<input type="checkbox"/> 2087268	NR	DEC	LOEC	REP	FCND	NR	SIG	42 d			
<input type="checkbox"/> 2087269	NR	DEC	NOEC	REP	FCND	NR	NOSIG	42 d			

**Figure 9. Companion Endpoint tab**

This tab allows users to link NOxx, LOxx and MATC values together.

To link the endpoints together, select the result that is to be linked from the result information view. Next, on the Companion Endpoint tab, select the result (s) that should be grouped together by marking the check box in front of the result. The Toxicity Module will perform an immediate check to ensure that the results can be linked together. If there is an error, the Toxicity Module will report the error for immediate rectification.

Note that all results that are to be linked must have the same result information, i.e. Duration, Response Site, Measurement, etc. A maximum of three (3) results records can be linked together, a NOxx, LOxx and MATC.

Example 1: The NOEC, LOEC and MATCs are reported for Growth - Length, Growth - Weight and Mortality - Mortality, the Growth Length endpoints are companion endpoints group 1, the Growth Weight are companion endpoints group 2 and the Mortality Endpoints are companion endpoints group 3. The data is coded as:

Endpoint: NOEC Effect: GRO Measurement: LGTH

Endpoint: LOEC Effect: GRO Measurement: LGTH Endpoint: MATC Effect: GRO Measurement: LGTH

Endpoint: NOEC Effect: GRO Measurement: WGHT Endpoint: LOEC Effect: GRO Measurement: WGHT

Endpoint: MATC Effect: GRO Measurement: WGHT

Endpoint: NOEC Effect: MOR Measurement: MORT

Endpoint: LOEC Effect: MOR Measurement: MORT

Endpoint: MATC Effect: MOR Measurement: MORT

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Attachment 1. ECOTOX Replicate Review Form

☐

Replicate  
Review  
Complete

ECOTOX 10% REPLICATE REVIEW

Reference Number: \_\_\_\_\_

Section I

Coordinator Comments (Data abstraction discrepancies outlined):

Initials:\_\_\_\_\_ Date:\_\_\_\_\_

Section II

Second Data Abstractor Comments (Response to discrepancies):

Initials:\_\_\_\_\_ Date:\_\_\_\_\_

Section III

First Data Abstractor Comments (Response/Incorporation of modifications):

Initials:\_\_\_\_\_ Date:\_\_\_\_\_

Section IV (If non-applicable, go to Section VI)

Coordinator Comments (Remaining data abstraction discrepancies outlined):

☐ Further action required, see below:

Initials:\_\_\_\_\_ Date:\_\_\_\_\_

Section V (If Section IV is used, complete Section V)  
First Data Abstractor Comments (Response/Incorporated modifications outlined):

Initials:\_\_\_\_\_ Date:\_\_\_\_\_

**Section VI**

☐ Replicate review complete; no further action required.